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UNITED STATES DISTRICT COURT

FOR THE CENTRAL DISTRICT OF CALIFORNIA

EASTERN DIVISION

UNITED STATES OF AMERICA, Plaintiff, v. CALIFORNIA STEM CELL TREATMENT CENTER, INC., *et al.* Defendants.	No. 5:18-CV-01005-JBG-KKx Hon. Jesus G. Bernal Riverside, Courtroom 1 **DECLARATION OF LARISSA LAPTEVA, M.D.** Hearing Date: August 5, 2019 Trial Date: October 1, 2019

I, Larissa Lapteva, hereby declare as follows:

1. I am the Associate Director in the Division of Clinical Evaluation and Pharmacology/Toxicology (DCEPT) in the Office of Tissues and Advanced Therapies (OTAT) in the Center for Biologics Evaluation and Research (CBER), at the United

States Food and Drug Administration (FDA). I received my medical degree (M.D.) from Moscow Medical Academy in 1997, my Master of Health Sciences (M.H.S.) degree from Duke University in 2006, and my Master of Business Administration (M.B.A.) degree from the University of Maryland in 2015. I am licensed to practice medicine in the State of Maryland and board-certified in Internal Medicine and Rheumatology (subsequently re-certified in Rheumatology). In addition to my current work at FDA, I serve as an attending physician/affiliated faculty at the National Institute of Arthritis, Musculoskeletal, and Skin Diseases (NIAMS) at the National Institutes of Health (NIH) where I provide clinical care to patients and teach rheumatology fellows and resident-physicians in training.

2. I have been employed by FDA since 2006. Since January 2017, I have served in my current position as the Associate Director of DCEPT, OTAT in CBER. In this role, I advise the Division Director of DCEPT and Office Director of OTAT and serve as a spokesperson for the Division and Office on medical and scientific aspects of various emerging, standing, complex, or precedent-setting regulatory and product-related issues and policy matters. In my current capacity, I direct projects, studies, and activities of concern to the Division and Office which often involve the coordinated effort of other Office, Center, or Agency components. Such projects may involve scientific or operational matters on cell, tissue, and gene therapies related policy, resulting from public health needs.

3. Prior to becoming the Associate Director in DCEPT/OTAT, I briefly served as the acting Division Director in the Division of Human Tissues in the same Office (OTAT) in the Center for Biologics Evaluation and Research. Prior to joining CBER in 2016, I worked in FDA's Center for Drug Evaluation and Research (CDER) between 2006 and 2016. During that period, I held various review and leadership positions (medical officer, team leader, division deputy director for safety, division director) which incorporated reviewing, recommending, and making decisions on development and approval of novel drugs, generic drugs, and biological products across a wide

range of therapeutic areas throughout all phases of product's lifecycle from early development to post-marketing. Between 2006 and 2011, I worked in the Office of New Drugs in CDER, first in the Division of Anesthesia, Analgesia, and Rheumatology Products (2006-2011), then in the Division of Pulmonary, Allergy, and Rheumatology Products (2011-2012), then in the Immediate Office of the Director in the Office of New Drugs coordinating activities across OND review divisions for applications with products aiming to treat rare diseases (2012-2014). In those roles, I reviewed and oversaw review activities related to numerous investigational new drug applications and new drug and biologic license applications (INDs, NDAs, and BLAs) as well as complex post-marketing issues for products regulated by the divisions. In 2014, following the passage of the Generic Drug User Fee Act, I was asked to serve as the Division Director in the Office or Research and Standards, Office of Generic Drugs in CDER. In that role (2014-2016), I led reorganization of Science Staff team into the Division of Therapeutic Performance (DTP), established the divisional infrastructure for processing of regulatory submissions, and planned, managed, organized, and directed all divisional activities, projects, and functions. I provided divisional level scientific and regulatory review and sign off on all controlled correspondence, pre-Abbreviated New Drug Application (pre-ANDA) submissions, responses to Citizens' Petitions, consults, product-specific bioequivalence guidances, thematic general guidances, white papers, and scientific articles prepared by the divisional staff.

4. Prior to joining FDA, I conducted clinical research at the National Institute of Dental and Craniofacial Research (NIDCR) and the National Institute of Arthritis, Musculoskeletal, and Skin Diseases (NIAMS) at the National Institutes of Health (NIH), providing clinical care to patients and leading clinical protocols including Phase 1-2 interventional studies in rheumatic conditions. In 2002, I completed Internal Medicine residency at the Forest Park Hospital (aff. St. Louis University) and in 2004, a Rheumatology fellowship at NIAMS, NIH.

5. By virtue of my training and professional experience, I am familiar with the

quantity and quality of evidence that is needed to establish the safety and effectiveness of drugs, the criteria for adequate and well-controlled clinical investigations, and also with the standards for evaluating whether a drug (or biological product) is "generally recognized ... as safe and effective" by qualified experts for its intended uses as set forth in the Federal Food, Drug, and Cosmetic Act (FDCA), 21 U.S.C. § 321(p). I am also familiar with the definition of "drug" as set forth in FDCA, 21 U.S.C. § 321(g)(1), with the definition of "biological product" as set forth in the Public Health Service Act (PHSA), 42 U.S.C. § 262(i), and with the definition of "prescription drug," under FDCA, 21 U.S.C. § 353(b)(1), as well as with the standards for determining whether a product is a prescription drug. Finally, I am familiar with the standard for evaluating the adequacy of directions for use of a drug product.

6. In preparing this declaration, I have conducted a thorough literature search and reviewed the declarations of Karlton Watson (Watson Decl.) and Carolyn Yong, Ph.D. (Yong Decl.), as well as certain evidence collected by investigators during FDA's inspections of the California Stem Cell Treatment Center (CSCTC), Inc. located in Beverly Hills (BH) and Rancho Mirage (RM), California (*see*, *infra*, ¶ 11).

7. As discussed in greater detail in Section I below, I conducted a review of whether a drug (and biological product) is generally recognized as safe and effective (GRAS/E) within the meaning of the FDCA for the three products manufactured and marketed by CSCTC. These products, as further described in this declaration below, are: (1) an autologous Stromal Vascular Fraction (SVF) product (hereinafter, "the SVF product"), (2) a culture-expanded SVF product [and/or adipose-derived stem cells—ADSCs] (hereinafter, "the expanded SVF product"), and (3) an SVF/Vaccinia product which combines the autologous SVF product with ACAM 2000®, Vaccinia Vaccine, Live (hereinafter, "the SVF/Vaccinia product"). The results of the literature search conducted to support this declaration and conclusions are provided in Attachment 1.

8. Additionally, I evaluated whether these products are prescription drugs within the meaning of the FDCA; this evaluation is provided in Section II of this

declaration.

9. Finally, I reviewed available limited information regarding the labeling of CSCTC products to evaluate the adequacy of the directions for use. My analysis is in Section III below.

Introduction

10. I reviewed the following records collected by FDA investigators during the July 17 through July 26, 2017 and July 21 through July 27, 2017 inspections of California Stem Cell Treatment Center, Inc. (CSCTC): Establishment Inspection Report of CSCTC's Rancho Mirage location (Watson Decl. Attachment 1); Establishment Inspection Report of CSCTC's Beverly Hills location (Watson Decl. Attachment 2); CSCTC promotional brochure (Watson Decl. Attachment 3). I also reviewed the following files that were downloaded from the internet: Cell Surgical Network (CSN) - CSCTC website screenshot 8-29-17 (Watson Decl. Attachment 7); and Paul Knoepfler Stem Cell Blog interviews with the Defendants (Watson Decl. Attachments 57, 58, 65). I also reviewed the Complaint for Permanent Injunction filed by the United States against CSCTC, CSN, Berman and Lander on May 9, 2018.

11. That evidence shows that CSCTC, located in Beverly Hills and Rancho Mirage, markets or has marketed the SVF product, the expanded SVF product, and the SVF/Vaccinia product to treat various diseases and conditions, which include but are not limited to: (a) for the autologous SVF product: arthritis, stroke, multiple sclerosis, traumatic brain injury, fibromyalgia, irritable bowel syndrome (IBS), optic neuropathy, spinal cord injury, Parkinson's disease, amyotrophic lateral sclerosis, chronic obstructive pulmonary disease; (b) for the expanded SVF product: Parkinson's disease, stroke, multiple sclerosis, amyotrophic lateral sclerosis, anoxic brain injury, hypothyroidism, Alzheimer's disease, diabetes, and renal failure; (c) for the SVF/Vaccinia product: different types of cancers, including advanced stage solid

tumors *(see* Watson Decl. Attachment 1 at 19). These products are administered by CSCTC through various routes including: intravenous, intraarticular, intrathecal, through the Ommaya reservoir, and via nebulization (*see* Watson Decl. Attachment 2 at 11). Although FDA understands that CSCTC no longer administers their SVF products to treat different ophthalmological conditions through the intravitreal route of administration, this review covers that intended use and the route as well because CSCTC used to administer these products intravitreally and now continues administering SVF through the following ocular routes of administration: "sub-tenons", "subconjunctival", "retrobulbar", and "topical" (*see* Watson Decl. Attachment 6 at 18).

12. The main subject product of this declaration, referred to as the SVF product, is a human cell, tissue, or cellular or tissue-based product (HCT/P) derived from a patient's own adipose tissue after recovery of the tissue following a liposuction procedure and subsequent processing of that tissue. Yong Decl. ¶¶ 19-20. The recovered adipose tissue is processed with enzymatic digestion, centrifugation, filtration, and other steps to obtain cellular components commonly referred to as SVF, which may be administered to the same individual from whom the adipose tissue was obtained. SVF may also be grown in culture for CSCTC by a third party located in Monmouth, New Jersey to produce the expanded SVF product. Yong Decl. ¶¶ 25-26. CSCTC's processes for manufacturing its various SVF products are discussed in more detail in the Yong Declaration. *See generally*, Yong Decl. ¶¶ 19-26. As set forth below, the SVF product is both a new drug under the FDCA and a biological product under the Public Health Service Act. Yong Decl. ¶¶ 13-14.

13. The other product described in this review that is herein referred to as SVF/Vaccinia product is also an HCT/P derived from a patient's adipose tissue that has been processed to yield an SVF product combined with reconstituted ACAM 2000®. ACAM 2000® is a live, Vaccinia Vaccine licensed in the U.S. for active immunization against smallpox disease for persons determined to be at high risk for smallpox

infection. CSCTC uses or has used the SVF/Vaccinia product for intravenous and intra-tumoral injection to patients diagnosed with different types of advanced cancers.

14. Based on my review of available scientific literature, I conclude that the overall contents of what is generally considered as SVF vary widely among patients, protocols, and studies, and depend largely on the intrinsic state of individual's tissues and procedures used for SVF isolation. The cell types found in any given SVF may include but may not be limited to the following: adipose tissue-derived stem cells (also referred to as mesenchymal stromal cells), pericytes, vascular adventitial cells, fibroblasts, pre-adipocytes, monocytes, macrophages, red blood cells, T-regulatory cells, mast cells, and smooth muscle cells.[1,2,3,4] The quantities and proportions of each cell type depend on individual patient's adipose tissue complexion, texture, and density as well as methods used to derive SVF, among other factors.[5] SVF culture expansion typically involves growth in culture and expansion of the obtained and enzymatically processed SVF cells using various reagents in the laboratory setting. The processes for SVF expansion and/or production of adipose-derived stem cells used by CSCTC's third-party manufacturer in New Jersey are not well described in the available materials which I reviewed.

15. The following potential effects of the different cell types found in SVF have been discussed in the literature: regenerative, secretory and excretory for various types

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[1] Zuk PA, et al; Multilineage cells from human adipose tissue: implications for cell-based therapies. Tissue Eng., 2001;7(2): 211-28.

[2] Zuk PA, et al; Human adipose tissue is a source of multipotent stem cells; Mol Biol Cell, 2002;13(12):4279-95.

[3] Mitchell, JB et al; Immunophenotype of human adipose-derived cells: temporal changes in stromal-associated and stem cell-associated markers. Stem Cells, 2006;24(2):376-385.

[4] Riordan NH et al; Non-expanded adipose stromal vascular fraction cell therapy for multiple sclerosis. J Transl Med. 2009; 7-29.

[5] Pak et al; Current use of autologous adipose tissue-derived stromal vascular fraction cells for orthopedic applications. J Biomed Science, 2017; 24(9);1-12.

of cytokines, chemokines, and growth factors, immunomodulatory, proangiogenic, anti-apoptotic, decreasing production of scar tissue, and anti-inflammatory.[6,7]

I. GRAS/E Analysis

GRAS/E Standard

16. Section 201(g)(1) of the FDCA provides that the term "drug" means: "(A) articles recognized in the official United States Pharmacopoeia, official Homoeopathic Pharmacopoeia of the United States, or official National Formulary, or any supplement to any of them; and (B) articles intended for use in the diagnosis, cure, mitigation, treatment, or prevention of disease in man or other animals; and (C) articles (other than food) intended to affect the structure or any function of the body of man or other animals; and (D) articles intended for use as a component of any articles specified in clause (A), (B), or (C). . .". 21 U.S.C. § 321(g)(1). As mentioned earlier, CSCTC markets their products to treat various diseases and conditions. Hence, by virtue of their use in treatment of various diseases and conditions and the expected potential to affect human body's structures and functions, the SVF product, the expanded SVF product, and the SVF/Vaccinia product are drugs within the meaning of FDCA, 201(g)(1). All three are also biological products. *See* Yong Decl. ¶¶ 13-14.

17. A drug (including one that is a biological product) is a "new drug" unless it is "generally recognized, among experts qualified by scientific training and experience to evaluate the safety and effectiveness of drugs, as safe and effective [(GRAS/E)] for use under the conditions prescribed, recommended, or suggested in the labeling". 21 U.S.C. § 321(p). At a minimum, the contention that a drug product is GRAS/E must be supported by the same quantity and quality of scientific evidence that is required to

[6] Pak et al; Current use of autologous adipose tissue-derived stromal vascular fraction cells for orthopedic applications. J Biomed Science, 2017; 24(9);1-12.

[7] Gimble JM, et al; Adipose-derived stem cells for regenerative medicine. Circ. Res 2007; 100:1249-1260.

obtain approval of a New Drug Application or Biologics License Application for the product.

18. A human drug (including a biological product) is GRAS/E if all of the following criteria are met:

A. The drug has been demonstrated to have at least "substantial evidence" of effectiveness for approval under 21 U.S.C. § 355(d), including adequate and well-controlled studies, *see Weinberger v. Hynson, Westcott and Dunning, Inc.*, 412 U.S. 609, 629-31; cf. 21 C.F.R. § 314.126;

B. The studies must be published in the scientific literature so that they are made generally available to the community of qualified experts; and

C. There must be a consensus of opinions by qualified experts, which is based on the published studies, that the drug is safe and effective for its labeled indications.

See 21 U.S.C. § 321(p)(1).

19. FDA's criteria for "adequate and well-controlled" studies are described in the Code of Federal Regulations and include, among other things, the following[8]:

A. There is a clear statement of the objectives of the investigation and a summary of the proposed or actual methods of analysis in the protocol for the study and in the report of its results. The protocol should contain a description of the proposed methods of analysis, and the study report should contain a description of the methods of analysis ultimately used. If the protocol does not contain a description of the proposed methods of analysis, the study report should describe how the methods used were selected.

B. The study must use a design that permits a valid comparison with a control to provide a quantitative assessment of the drug effect. The protocol for the study and report of results should describe the study design precisely; for example, duration of treatment periods, whether treatments are parallel,

[8] 21 CFR § 314.126(b).

sequential, or crossover, and whether the sample size is predetermined or based upon some interim analysis.

C. The method of selection of subjects provides adequate assurance that they have the disease or condition being studied, or evidence of susceptibility and exposure to the condition against which prophylaxis is directed.

D. The method of assigning patients to treatment and control groups minimizes bias and is intended to assure comparability of the groups with respect to pertinent variables such as age, gender, severity of disease, duration of disease, and use of drugs or therapy other than the test product. The protocol for the study and the report of its results should describe how subjects were assigned to groups. Ordinarily, in a concurrently controlled study, assignment is by randomization, with or without stratification.

E. Adequate measures are taken to minimize bias on the part of the subjects, observers, and analysts of the data. The protocol and report of the study should describe the procedures used to accomplish this, such as blinding.

F. The methods of assessment of subjects' response are well-defined and reliable. The protocol for the study and the report of results should explain the variables measured, the methods of observation, and criteria used to assess response.

G. There is an analysis of the results of the study adequate to assess the effects of the drug. The report of the study should describe the results and the analytic methods used to evaluate them, including any appropriate statistical methods. The analysis should assess, among other things, the comparability of test and control groups with respect to pertinent variables, and the effects of any interim data analyses performed.

21. Reports of adequate and well-controlled investigations provide the primary basis for determining whether there is "substantial evidence" to support the claims of effectiveness for new drugs and biological products. Therefore, the study report should

provide sufficient details of study design, conduct, and analysis to allow critical evaluation and a determination of whether the characteristics of an adequate and well-controlled study are present.[9]

22. For a study to be considered adequate for approval of a new drug or a biological product, it is also required that the test product be standardized as to identity, strength, quality, purity, and dosage form to give significance to the results of the investigation.[10]

23. Confirmatory studies are generally necessary to provide firm evidence of efficacy or safety.[11] A confirmatory study is an adequately controlled trial in which the hypotheses are stated in advance and evaluated. In such studies, the key hypothesis of interest follows directly from the trial's primary objective, is always predefined, and is the hypothesis that is subsequently tested when the study is complete. In a confirmatory study, it is equally important to estimate, with due precision, the size of the effects attributable to the treatment of interest and to relate these effects to their clinical significance.[12]

24. Uncontrolled studies or partially controlled studies are not acceptable as the sole basis for the approval of claims of effectiveness.[13] Such studies carefully conducted and documented, may provide corroborative support of well-controlled studies regarding efficacy and may yield valuable data regarding safety of the test drug.[14] Such studies will be considered on their merits in the light of the principles

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[9] *See* 21 CFR § 314.126(a).

[10] *See* 21 CFR § 314.126(d).

[11] Guidance for Industry: E9 Statistical Principles for Clinical Trials; September 1998, at 4-5.

[12] *Id.*

[13] *See* 21 C.F.R. § 314.126(e).

[14] *Id.*

listed in 21 C.F.R. § 314.126, with the exception of the requirement for the comparison of the treated subjects with controls.[15] Isolated case reports, random experience, and reports lacking the details which permit scientific evaluation will not be considered.[16]

25. To assess whether there is clinical literature to support a determination of GRAS/E, and to identify any adequate and well-controlled clinical studies or trials of safety and effectiveness of the three aforementioned products, I conducted a search of established medical literature databases, specifically, PubMed, EMBASE, Web of Science, and Cochrane [Clinical Trials], for the three products and various selected diseases and conditions that these products are intended to treat. *See, supra*, ¶ 11.

Databases Evaluated for this Declaration

26. For the search conducted to support this declaration, I used the following search terms within each of the aforementioned databases:

(a) "stromal vascular fraction", "SVF", "adipose-derived stem cells"; each term was paired with each of the following medical conditions: arthritis, stroke, multiple sclerosis, traumatic brain injury, fibromyalgia, irritable bowel syndrome, optic neuropathy, spinal cord injury, Parkinson's disease, amyotrophic lateral sclerosis, and chronic obstructive pulmonary disease;

(b) "expanded stromal vascular fraction", "ATCELL"; each term was paired with each of the following medical conditions: hypothyroidism, stroke, arthritis, Alzheimer's disease, multiple sclerosis, Parkinson's disease, renal failure, amyotrophic lateral sclerosis, anoxic brain injury, and diabetes;

(c) "stromal vascular fraction and vaccinia", "stromal vascular fraction and ACAM 2000"; each term was paired with the word "cancer".

I summarize the searches below. The results of the searches are documented in Attachment 1 to this declaration.

[15] 21 C.F.R. § 314.126(e).

[16] *Id.*

27. **EMBASE** is an abstract and indexing database specialized in the biomedical field and pharmaceutical research, with power searching tools enabling researchers to retrieve essential biomedical information. It covers the international biomedical literature from 1947 to the present day.

Search Limits: Years 1-1-1990 through 11-20-2017; included Cochrane Review, Systematic Review, Controlled Clinical Trial, Randomized Controlled Trial.

Search Dates: 11-20-2017 and 11-24-2017; updated on 10-23-2018 and 04-03-2019.

28. **Web of Science** is a multidisciplinary resource for searching, accessing, and analyzing journal literature. It provides a comprehensive citation search and covers over 90 million records from scientific journals indexed from as early as 1900 to the present.

Search Limits: No limits.

Search Date: 11-26-2017; updated on 10-23-2018 and 04-03-2019.

29. **Cochrane Central Register** [Clinical Trials] is a bibliography of clinical trials identified by contributors to the Cochrane Collaboration and others, as part of an international effort to search the world's journals and create an unbiased source of data for systematic reviews and trial information. The database is also linked to trials listed at https://clinicaltrials.gov/

Search Limits: No limits.

Search Date: 11-27-2017; updated on 10-23-2018 and 04-03-2019.

30. **PubMed** is produced by the U.S. National Library of Medicine and currently comprises over 29 million citations for biomedical literature from MEDLINE, life science journals, and online books. PubMed citations and abstracts include the fields of biomedicine and health, covering portions of the life sciences, behavioral sciences, chemical sciences, and bioengineering.

Search Limits: Clinical trial, human.

Search Dates: 12-11-2017 and 12-12-2017; updated on 10-22-2018 and 04-03-2019.

31. The results of the search of these four databases are presented in

Attachment 1. As noted earlier, to be considered GRAS/E, a drug (or biological product) must be demonstrated to have at least "substantial evidence" of effectiveness for approval under 21 U.S.C. § 355(d), including adequate and well-controlled studies as defined in 21 C.F.R. § 314.126. For the three products intended for the treatment of the listed diseases and conditions, such adequate and well-controlled studies are clinical studies conducted in humans. Therefore, for the purposes of this declaration, all animal studies and studies describing in-vitro investigations have been excluded from the listings shown in Attachment 1.

Discussion of GRAS/E criteria

32. The search of published clinical studies revealed that none of the three products of interest has demonstrated substantial evidence of effectiveness for the listed diseases and conditions; nor has any of the three been studied in adequate and well-controlled clinical studies as defined in 21 C.F.R. § 314.126. The vast majority of the studies published in the scientific literature, while made generally available to the community of qualified experts, comprised of in-vitro experiments and preclinical animal model investigations. The limited amount of studies that were conducted in humans with different disease populations were primarily case-report series, or uncontrolled studies conducted without properly blinded, statistically prespecified comparative assessments, or with investigational treatments that combined the products of interest and other components such as, for example, platelet-rich plasma, and lacked one or more of the required elements of design that would make them to be considered as adequate and well-controlled (*see* Attachment 1). Therefore, the three products described in this declaration have not been demonstrated to have at least "substantial evidence" of effectiveness for approval under 21 U.S.C. § 355(d),[17] based on adequate and well-controlled studies as defined in 21 C.F.R. § 314.126.

33. A number of qualified experts from different therapeutic areas, including

[17] *See also* 21 U.S.C. § 262 (establishing criteria for approval of a biologics license application).

treatment of stroke,[18] amyotrophic lateral sclerosis, Parkinson's disease,[19] spinal cord injury,[20,21] chronic obstructive pulmonary disease,[22,23] eye disease [24] osteoarthritis [25]and diabetes[26,27,28] among others, published their opinions and assessments of the current state of investigations related to the treatments with stem cells, including autologous adipose tissue-derived stem cells and stromal vascular fraction. While many experts proposed that there is a good potential in use of these cell-based products for various therapeutic indications, it was noted in different expert opinion articles across a wide range of medical disciplines that the current scientific evidence is rooted mainly in the in-vitro experiments and animal studies, hence resulting in the conclusion that

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[18] Chan TM et al; The use of ADSCs as a treatment for chronic stroke. Cell Transplant. 2014;23(4-5):541-7

[19] Chang, KA et al; Therapeutic Potential of Human Adipose-Derived Stem Cells in Neurological Disorders. J Pharm Sciences, Dec 2014; 126(4); 293-301

[20] Xiang LB et al; Stem cell transplantation for treating spinal cord injury. A literature comparison between studies of stem cells obtained from various sources. Neural Regeneration Research, 2012;7(16), 1256-1263

[21] Shende P, Subedi M; Pathophysiology, mechanisms and applications of mesenchymal stem cells for the treatment of spinal cord injury. Biomedicine and Pharmacotherapy, 2017 Jul;91:693-706

[22] Wecht, S et al; Mesenchymal stem cells in the treatment of chronic lung disease; Respirology, 2016, 21(8),1366-1375

[23] Oh DK, et al; Lung regeneration therapy for chronic obstructive pulmonary disease. Tuberculosis and Respiratory Diseases Jan 2017, 80:1 (1-10) 1

[24] Mead B et al; Stem cell treatment of degenerative eye disease. Stem Cell Res. 2015 May;14(3):243-57

[25] Pak J, et al; Cartilage regeneration in human with adipose tissue-derived stem cells and adipose stromal vascular fraction cells: Updated Status. Int. J Mol. Sci. Jul 2018, 19(7): 2146.

[26] Nguyen A, Guo J, et al; Stromal vascular fraction: A regenerative reality? Part 1: Current concepts and review of the literature. JPRAS, Feb 2016, 69(2), 170-179

[27] Guo J, Nguyen A et al; Stromal vascular fraction: A regenerative reality? Part 2: Mechanisms of regenerative action. JPRAS, Feb 2016, 69(2), 180-8

[28] Gharravi, AM, Jafar A, et al; Current status of stem cell therapy and scaffolds for the treatment of diabetes mellitus. Diabetes and Metabolic Syndrome: Clinical Research and Reviews 2018 12:6 (1133-1139)

appropriately conducted clinical trials are needed to adequately evaluate safety and effectiveness of these products in different diseases. In November 2015, the American Academy of Orthopedic Surgeons (AAOS) conducted a symposium on biologic treatments (i.e., adipose tissue-derived stem cells and other stem cell-based therapies as well as scaffold-based treatments) of orthopedic injuries and subsequently published a consensus report on the topic identifying significant knowledge gaps and the need for future research in appropriately designed clinical trials evaluating these therapies.[29]

34. In addition, significant safety concerns were raised in clinical literature describing serious adverse occurrences of retinal detachments, increased intraocular pressure, vitreous hemorrhage, and vision loss[30,31] that resulted from intravitreal administration of autologous adipose tissue-derived stem cells, and serious events of pulmonary embolism occurring after intravenous administration of autologous adipose tissue-derived stem cells.[32] Following some of these reports, the American Academy of Ophthalmology[33] and the American Lung Association[34] issued warnings to consumers about the potential for harm and the lack of proven benefit of these treatments. Thus, there is no consensus of opinions by qualified experts that the products described in this declaration are safe and effective for their labeled indications. (*See* also Section III below).

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[29] LaPrade R, et al; AAOS Research symposium updates and consensus: biologic treatment of orthopedic injuries. J Am. Acad. of Orthop. Surg., 2016; 24: e62-e78.

[30] Kuriyan AE et al; Vision Loss after Intravitreal Injection of Autologous "Stem Cells" for AMD. N Engl. J Med. 2017 Mar 16;376(11):1047-1053

[31] Saraf SS, et al. Bilateral Retinal Detachments After Intravitreal Injection of Adipose-Derived 'Stem Cells' in a Patient with Exudative Macular Degeneration. Ophthalmic Surg. Lasers Imaging Retina. Sep. 2017;48(9): 772-775.

[32] Jung JW et al; Familial occurrence of pulmonary embolism after intravenous adipose tissue-derived stem cell therapy. Yonsei Med J, 2013;54: 1239-96.

[33] American Academy of Ophthalmology Statement on Stem Cell Therapy for Treating Eye Disease https://www.aao.org/newsroom/news-releases/detail/statement-stem-cell-therapy-treatment-eye-disease

[34] Stem Cell Therapy for Lung Diseases https://www.lung.org/our-initiatives/research/about-our-research/stem-cell-therapy.html

35. In summary, based on my review of the scientific and clinical literature, none of the products that are subject of this declaration, namely the SVF product, the expanded SVF product, and the SVF/Vaccinia product meets all three criteria for GRAS/E described in Section I.

GRAS/E conclusion

36. I conclude, based on my professional experience and the conducted literature search, that

A. the SVF product **IS NOT** GRAS/E;

B. the expanded SVF product **IS NOT** GRAS/E; and

C. the SVF/Vaccinia product **IS NOT** GRAS/E.

II. Prescription Drug Analysis

37. A prescription drug is a drug intended for use by man which, because of its toxicity or other potentiality for harmful effect, or the method of its use, or the collateral measures necessary to its use, is not safe for use except under the supervision of a practitioner licensed by law to administer such drug; or a drug limited by an approved application under Section 505 of the Act (21 U.S.C. § 355) for use under the professional supervision of a practitioner licensed by law to administer such drug.[35]

38. CSCTC's SVF and expanded SVF products are administered by CSCTC through various routes including: intravenous, intraarticular, intrathecal, intradermal, through the Ommaya reservoir, and via nebulization (the SVF product); and intravenous, intraarticular, and intraventricular (the expanded SVF product). The SVF/Vaccinia product is administered intravenously and by direct injection into the cancer lesions. *See* Watson Decl. Attachment 2 at 11-12.

39. Although FDA understands that CSCTC no longer markets their SVF and expanded SVF products to treat ophthalmological conditions through the intravitreal route of administration, CSCTC, under its protocol for autologous adipose tissue-

[35] 21 U.S.C. § 353(b)(1).

derived stromal vascular fraction "to repair ophthalmologic conditions" (*see* Watson Decl. Attachment 1 at 17; Attachment 5 at 2), continues to administer these products using the following routes of administration: "sub-tenons", "subconjunctival", "retrobulbar", and "topical" (*see* Watson Decl. Attachment 6).

40. The three products are used by CSCTC to treat a wide range of diseases or conditions, including the following (*see* Watson Decl. Attachment 2 at 11):

A. The SVF product – arthritis, stroke, multiple sclerosis, traumatic brain injury, fibromyalgia, irritable bowel syndrome, optic neuropathy, spinal cord injury, Parkinson's disease, amyotrophic lateral sclerosis, and chronic obstructive pulmonary disease;

B. The expanded SVF product – Parkinson's disease, stroke, multiple sclerosis, amyotrophic lateral sclerosis, anoxic brain injury, hypothyroidism, Alzheimer's disease, diabetes, and renal failure; and

C. The SVF/Vaccinia product – advanced stage cancers.

Therefore, all three products at issue meet the following criteria for classification as prescription drugs:

(1) Collateral measures necessary for safe, effective, and appropriate use:

a. Medical expertise, licensure, and appropriate subspecialty training are required to diagnose the diseases and condition(s) to be treated and to determine the appropriate therapeutic intervention(s) for the diseases and conditions listed above.

b. Medical expertise, licensure, and/or appropriate training are also required to administer the product through the intended parenteral routes of administration.

c. Medical expertise and training are required for the appropriate administration of Vaccinia Vaccine, live (ACAM 2000®) that is FDA

approved to be administered by the percutaneous route (scarification)[36], and not intravenously or into the cancer lesions, as per CSCTC's use. Also, Vaccinia Vaccine is contraindicated for administration to patients who have severe immunodeficiency. Notably, patients with advanced stage cancers are often severely immunocompromised due to their disease or treatments' side effects and, therefore, at a greater risk for development of complications associated with the use of Vaccinia Vaccine, live (ACAM 2000®).

(2) Toxicity or other potentiality for harmful effects:

a. Potential infections and other local and systemic reactions may be associated with administration of cell-based products when given intravenously and through other intended parenteral routes of administration. Several reports from the available literature describe adverse occurrences of administration site reactions such as swelling, tendonitis[37], and intra-articular pain[38], as well as systemic reactions manifested by transient fever,[39] facial flushing and myalgia,[40] and pulmonary embolism.[41,42] Intravitreal administration of autologous adipose-derived stem cells has resulted in retinal detachments,

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[36] Vaccinia Vaccine, Live; Prescribing Information: https://www.fda.gov/downloads/BiologicsBloodVaccines/Vaccines/ApprovedProducts/UCM142572.pdf

[37] Pak J et al; Safety reporting on implantation of autologous adipose tissue-derived stem cells with platelet-rich plasma into human articular joints. BMC, Musculoskelet Disord, 2013;14-337

[38] Siennicka K et al; Adipose-derived cells (stromal vascular fraction) transplanted for orthopedic or neurological purposes: are they safe enough? Stem Cell International, 2016 Article ID 5762916

[39] Lalu MM et al; Safety of cell therapy with mesenchymal stromal cells (SafeCell): a systematic review and meta-analysis of clinical trials. PLoS ONE 2012;7 (10): e47559

[40] Rodriguez JP et al; Autologous stromal vascular fraction therapy for rheumatoid arthritis: rationale and clinical safety. Int. Arch Med, 2012, 5:5

[41] Jung JW et al; Familial occurrence of pulmonary embolism after intravenous adipose tissue-derived stem cell therapy. Yonsei Med J, 2013;54: 1239-96.

[42] Tatsumi et al; Tissue factor triggers procoagulation in transplanted mesenchymal stem cells leading to thromboembolism. Biomedical and Biophysical Research Communications 2013, 431; 203-209

increased intra-ocular pressure, vitreous hemorrhage, and vision loss.[43,44]

b. Potentially serious neurological complications may be associated with the products' intended routes of administration, including through the Ommaya reservoir and intrathecally. Other occurrences reported with intrathecal administration of autologous SVF included temporary low back and leg pain associated with elevated cerebrospinal fluid protein and nucleated cells with MRI imaging demonstrating thickened lumbosacral nerve roots,[45] and urinary tract infection, headache, nausea, and vomiting.[46]

c. Serious adverse reactions described in the boxed warning of Vaccinia Vaccine, live (ACAM2000®) are associated with administration of Vaccinia Vaccine, live (ACAM2000®) which include but are not limited to: myocarditis, pericarditis, encephalitis, encephalomyelitis, encephalopathy, progressive and generalized vaccinia, severe vaccinial skin infections, erythema multiforme major, including Stevens-Johnson syndrome, ocular complications, blindness, eczema vaccinatum resulting in permanent sequalae or death.[47]

41. Based on the collateral measures necessary for the use of these products,

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[43] Kuriyan AE et al; Vision Loss after Intravitreal Injection of Autologous "Stem Cells" for AMD. N Engl. J Med. 2017 Mar 16;376(11):1047-1053

[44] Saraf SS, et al. Bilateral Retinal Detachments After Intravitreal Injection of Adipose-Derived 'Stem Cells' in a Patient with Exudative Macular Degeneration. Ophthalmic Surg. Lasers Imaging Retina. Sep. 2017;48(9): 772-775.

[45] Staff N et al; Safety of intrathecal autologous adipose-derived mesenchymal stromal cells in patients with ALS. Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration; 2016; 17 Supplement 1 (242)

[46] Hur, JW et al; Intrathecal transplantation of autologous adipose-derived mesenchymal stem cells for treating spinal cord injury: A human trial. Journal of Spinal Cord Medicine; SN 1079-0268, 2016 (39) 6:655-664

[47] Vaccinia Vaccine, Live; Prescribing Information: https://www.fda.gov/downloads/BiologicsBloodVaccines/Vaccines/ApprovedProducts/UCM142572.pdf

their expected toxicities and potential for harmful effects, I conclude that these three products cannot be self-administered safely by lay people and, if administered, should be administered only under the supervision of a practitioner licensed by law to administer such products.

42. In addition, the diseases and conditions for which CSCTC is currently marketing the products of interest cannot be self-diagnosed, and these diseases and conditions are not amenable to self-treatment.

Prescription drug conclusion

43. Therefore, based on my training and professional experience, I conclude that due to the potential and known toxicities that may occur with their use, the method of their use and the collateral measures necessary to their use, the three products, i.e., the SVF product, the expanded SVF product, and the SVF/Vaccinia product are prescription drugs within the meaning of 21 U.S.C. § 353(b).

III. Directions for Use

44. Drug labeling refers to all the printed information that accompanies a drug or biological product, including the wrapping and package insert. Drug labeling is regulated by FDA. *See* 21 U.S.C. § 352; *see also* 21 U.S.C. § 355(a), (d); 21 C.F.R. Part 201.

45. The FDCA requires the labeling of a drug to bear "adequate directions for use," and FDA, by regulation, has defined adequate directions for use to mean "directions under which the layman can use a drug safely and for the purposes for which it is intended." *See* 21 U.S.C § 352(f)(1) and 21 C.F.R. § 201.5. Directions for use may be inadequate unless the label contains, among other things, statements of: all conditions, purposes, or uses for which the drug is intended; quantity of dose, including, among others, the usual quantities for each of the uses for which it is intended and usual quantities for persons of different physical conditions; frequency and duration of administration; time of administration in relation to time of meals,

onset of symptoms, and other factors; and route or method of administration. *See* 21 C.F.R. § 201.5(a)-(g).

46. Some drugs are regulated as prescription drugs, meaning they can be dispensed only by valid prescription. As discussed above, CSCTC's products are prescription drugs under the FDCA. *See, supra*, Section II.

47. A prescription drug may be dispensed only pursuant to a physician's prescription and, by its nature, may be used safely only under the supervision of a physician. *See* 21 U.S.C. § 353(b)(1)(B). Prescription drugs for human use are exempt from the requirement that their labeling bear adequate directions for use if certain criteria are met, including that the labeling on or within the package from which the drug is to be dispensed contains "adequate information for use," including "indications, effects, dosages, routes, methods, and frequency and duration of administration, and any relevant hazards, contraindications, side effects, and precautions under which practitioners licensed by law to administer the drug can use the drug safely and for the purposes for which it is intended, including all purposes for which it is advertised or represented." 21 C.F.R. § 201.100.

48. CSCTC's staff in the Beverly Hills facility has stated that there is no labeling for their products that are both manufactured and administered at CSCTC BH. If SVF is transported to different locations, the container is typically labeled with the patient's name, date of birth, date processed, and the staff's initials (*see* Watson Decl. Attachment 2 at 19). At CSCTC facility in Rancho Mirage, the final product labeling is represented by hand-written patient's last name and first initial and the date when SVF is "to be deployed" (*see* Watson Decl. Attachment 1 at 33). There is no written procedure for final product labeling in either facility. No other information is provided: there is no information about, for example, indications, dosages, frequency and duration of administration, or route or method of administration. The only product that does have an adequate label is the Vaccinia Vaccine component of the SVF/Vaccinia product. However, as described above, CSCTC does not use the Vaccinia Vaccine live

(ACAM2000®) according to its approved dosing, the labeled indication, and the labeled instructions for safe use. As such, the labeling of any of CSCTC's products does not contain adequate information for use and does not satisfy the standard for adequate directions for use, as per 21 C.F.R. § 201.5, or per 21 C.F.R. § 201.100.

49. Even if CSCTC attempts to label their products with adequate information and directions for use, it is currently not possible to do so. As noted above, adequate information and directions for use presented in a product's labeling must include, among other things, specific indications, dose and route of administration, warnings, precautions, contraindications, adverse reactions, and other information pertinent to safe and effective use of the product. To this end, the labeling of a drug product must contain information that can be obtained from well-designed and properly conducted scientific and clinical investigations (including adequate and well-controlled studies) that would enable sound scientific conclusions about the drug's safety and effectiveness. As discussed above, *see supra*, Section I, there are no sufficient scientific data on which adequate information or adequate directions for using of any of the three products described herein could possibly be based.

50. As discussed above, CSCTC's products do not currently have adequate labeling. In addition, the studies published in the available literature are not adequate and well-controlled, as per 21 C.F.R. § 314.126, and no adequate data regarding safety and substantial effectiveness of these products for the aforementioned diseases and conditions can be obtained from these studies. Therefore, at this time, none of CSCTC's products can possibly satisfy the requirement that its labeling bear adequate directions for use.

Directions for Use Conclusion

51. Based on the information presented above, I conclude that, at present, labeling for any of the CSCTC products does not – and cannot – bear adequate directions for use.

IV. Additional Support for Other Potentiality for Harmful Effects

52. Establishment Inspection Reports at CSCTC's Beverly Hills and Rancho Mirage facilities (*see, generally,* Watson Decl. Attachments 1 and 2) described the lack of adequate procedures and the absence of functional infrastructure for appropriate medical monitoring of patients with serious and chronic systemic conditions (*see* Section II) after administration of CSCTC's products. While assessing these observations was not the primary subject of this declaration, it is worthwhile to note one particularly egregious inspectional observation which concerns 25 patients with advanced cancers who were treated with the SVF/Vaccinia product. According to the EIR report, following the SVF/Vaccinia product administration, at least 8 patients were described to develop itchy rashes and blisters of variable severity and extent as well as other occurrences (*see* Watson Decl. Attachment 1 at 21-24). It is notable that the records are incomplete, and the occurrences are not fully documented. Such reactions in the severely immunocompromised patients with advanced cancers likely represented vaccinia infection and post-administration adverse reactions occurring after the SVF/Vaccinia product administration and manifesting as an eruption of skin lesions caused by vaccinia. Vaccinia Vaccine live (ACAM2000®) labeling contains a boxed warning describing the expected adverse reactions including serious skin and systemic reactions[48] (*see* Section II). This vaccine is contraindicated for administration to patients with severe immunodeficiencies (including patients with acquired immunodeficiency states associated with malignancies—Sections 4.0 and 5.4 Vaccinia Vaccine live (ACAM2000®) Prescribing Information)[49] because these patients are not expected to benefit from the vaccine and are at high risk for developing a clinical presentation of severe vaccinia infection as well as other adverse reactions associated with the live Vaccinia Vaccine. Further, according to the inspectional description, most

[48] Vaccinia Vaccine, Live; Prescribing Information: https://www.fda.gov/downloads/BiologicsBloodVaccines/Vaccines/ApprovedProducts/UCM142572.pdf

[49] *Id.*

patients died within 1-3 months of CSCTC's product administration (*see* Watson Decl. Attachment 1 at 49). Given the limited information provided in the report, it is unclear to which extent, if any, the administration of CSCTC's product may have contributed to the lethal outcomes in these patients.

53. Patients with advanced cancers, in addition to having regional complications of locally expanded and metastasized tumors, often suffer from continuous pain requiring analgesics, chronic intractable malaise, and sometimes cognitive decline, among other disease manifestations. Making their already distressed condition worse by inducing the expected adverse occurrences in this patient population through administration of the live Vaccinia Vaccine in the face of the unlikely benefit from the given SVF/Vaccinia experimental treatment cannot be medically and ethically justified.

I declare under penalty of perjury that the foregoing is true and correct. 28 U.S.C. § 1746.

Executed on June 24, 2019.

Larissa Lapteva, M.D.
Associate Director
U.S. Food and Drug Administration
Center for Biologics Evaluation and Research
Office of Tissues and Advanced Therapies
Division of Clinical Evaluation, Pharmacology, and Toxicology

Attachment 1: Search Results

EMBASE

Arthritis

(1) Comparative clinical observation of arthroscopic microfracture in the presence and absence of a stromal vascular fraction injection for osteoarthritis.

Nguyen P.D., Tran T.D.-X., Nguyen H.T.-N., Vu H.T., Le P.T.-B., Phan N.L.-C., Vu N.B., Phan N.K., van Pham P.

Stem Cells Translational Medicine, Jan 2017 6:1 (187-195).

ABSTRACT: Osteoarthritis (OA) is a degenerative cartilage disease that is characterized by a local inflammatory reaction. Consequently, many studies have been performed to identify suitable prevention and treatment interventions. In recent years, both arthroscopic microfracture (AM) and stem cell therapy have been used clinically to treat OA. This study aimed to evaluate the clinical effects of AM in the presence and absence of a stromal vascular fraction (SVF) injection in the management of patients with OA. Thirty patients with grade 2 or 3 (Lawrence scale) OA of the knee participated in this study. Placebo group patients (n = 15) received AM alone; treatment group patients (n = 15) received AM and an adipose tissue-derived SVF injection. The SVF was suspended in platelet-rich plasma (PRP) before injection into the joint. Patient groups were monitored and scored with the Western Ontario and McMaster Universities Arthritis Index (WOMAC), Lysholm, Visual Analog Pain Scale (VAS), and modified Outer bridge classifications before treatment and at 6, 12, and 18 months post-treatment. Bone marrow edema was also assessed at these time points. Patients were evaluated for knee activity (joint motion amplitude) and adverse effects relating to surgery and stem cell injection. Treatment efficacy was significantly different between placebo and treatment groups. All treatment group patients had significantly reduced pain and WOMAC scores, and increased Lysholm and VAS scores compared with the placebo group. These findings suggest that the SVF/PRP injection efficiently improved OA for 18 months after treatment. This study will be continuously monitored for additional 24 months.

(2) Bone marrow concentrated cells and stromal vascular fraction cells injections for osteoarthritis treatment: A systematic review.

Vadalà G., Papalia R., Verde L.L.A., Russo F., Denaro V., Rosa M.A.

Journal of Biological Regulators and Homeostatic Agents Sep 2016; 30:4 Supplement 1 (69-76);

ABSTRACT: The aim of this systematic review is to examine current clinical evidence supporting the intraarticular injection of bone marrow concentrate cells (BMC) and adipose-

derived stromal vascular fraction cells (SVF) for the treatment of osteoarthritis (OA). The research was performed on PubMed (Medline), EMBASE and Cochrane Library considering the English literature. Only clinical trials have been included. The systematic research identified twelve clinical trials. Articles included in the study, were one of level II, four of level III, six of level IV and one level V. Among clinical trials, none were randomized, four were comparative, seven were case series, and one was a case report. Seven studies were focused on the use of SVF (1332 patients) and five on the use of BMC (963 patients), with preliminary interesting findings in the OA treatment. Despite the growing interest in this biological approach for OA, knowledge on this topic is still preliminary. Randomized controlled trials are needed to support the potential of BMC and SVF injections and to evaluate advantages and disadvantages with respect to the available treatments.

(3) A controlled clinical trial of adipose derived stem cell transplantation for osteoarthritis.

Pham P.V.

Cytotherapy, June 2016; 18:6 SUPPL. 1 (S12-S13).

ABSTRACT: Adipose-derived stem cell (ADSC) transplantation is a promising therapy for some chronic diseases. Some previous clinical trials showed that some diseases could improve and recover by ADSC transplantation. This study aimed to investigate ADSC transplantation effects on osteoarthritis compared to control. This clinical trial was performed at 3 hospitals with controlled groups. There were total 60 patients enrolling this study. All patients were divided into 3 groups: Group 1 (15 patients, traditional treatment therapy by endoscopic surgery- as control), Group 2 (15 patients; endoscopic surgery + ADSC injection), Group 3 (ADSC injection). In the Group 2, patients were injected with an autologous mixture of stem cell-enriched fractions (stromal vascular fraction-SVF) and activated platelet rich plasma (aPRP) that were prepared from fat tissue, and peripheral blood respectively, while in group 3, patients only were injected with SVF and aPRP without endoscopic surgery. The results showed that all patients in treated groups significantly reduced pain, reduced the WOMAC score, clearly increased the Lysholm scores and VAS scores compared to the control group after 18 months. These findings suggested injection of SVFs and a PRP mixture efficiently improved the osteoarthritis after six months.

(4) Safety of stromal vascular fraction cells applications in chronic pain.

Lander E.B., Berman M.H., See J.R.

Techniques in Regional Anesthesia and Pain Management 2015 19 :1-2(10 - 13)

ABSTRACT: Autologous stromal vascular fraction (SVF) can be enzymatically released from lipoaspirate obtained under local anesthesia. SVF is known to have regenerative, anti-inflammatory, pain mitigating, and immune-modulatory properties. Our translational research

network has been studying the safety and efficacy of SVF since 2012. Almost 100 related physician teams around the world are applying the same institutional review board–approved methods of SVF production, which use a surgically closed SVF isolation system. During the same outpatient surgical procedure, procured SVF is administered according to strict investigative protocols to mitigate diseases associated with chronic pain including arthritis, autoimmune disease, neurodegenerative disease, and various inflammatory conditions. The shared research collaborative online database contains safety and efficacy data on more than 3500 patients. Our processed SVF contains valuable anti-inflammatory cytokine growth factors in addition to both adult mesenchymal and hematopoietic stem cells targeting damaged, or inflamed tissue. SVF administration may potentially play a large role in the outpatient treatment of pain. In this article, we describe our protocol for the production and administration of SVF, and its safety and efficacy in the treatment of pain associated with chronic conditions.

(5) A Prospective Safety Study of Autologous Adipose-Derived Stromal Vascular Fraction Using a Specialized Surgical Processing System.

Berman, M. and Lander, E.

The American Journal of Cosmetic Surgery 2017, Vol. 34(3) 129–142

ABSTRACT: Autologous adipose-derived stromal vascular fraction (SVF) has been proposed as a remedy for a number of inflammatory, autoimmune, and degenerative conditions. This procedure had mainly been evaluated in veterinary medicine and outside the United States when this study was initiated. This study looks at adverse events to evaluate safety as its primary and secondarily follows efficacy of SVF as deployed through intra-articular injections and intravenous infusions for a variety of orthopedic and non-orthopedic conditions. We hypothesized that autologous SVF deployment using a specialized surgical processing system (the CSN Time Machine® system, trademark name for the MediKhan Lipokit/Maxstem system; MediKhan, Los Angeles, California) was safe (i.e., minimally acceptable adverse events) and that clinical efficacy could be demonstrated. This was a prospective case series. After institutional review board approval, 1698 SVF deployment procedures were performed between 2011 and 2016 by us and other affiliates with our same system trained by us as a nearly closed sterile surgical lipotransfer procedure on 1524 patients with various degenerative, inflammatory, and autoimmune conditions with a majority involving the musculoskeletal system. All outcome test data were collected in an online database over a 5-year period. Our study shows a very low number of reported adverse events and a reduction in pain ratings after 6 months or more across a variety of musculoskeletal diseases and improvements in a variety of other degenerative conditions. Our system for producing adipose-derived SVF therapy for our patients was safe and benefits could be measured for a long time after SVF deployment. Further controlled long-term studies for specific disease conditions with large patient populations are necessary to further investigate the benefits observed.

Amyotrophic Lateral Sclerosis

(1) Safety of intrathecal autologous adipose-derived mesenchymal stromal cells in patients with ALS.

Staff N., Madigan N., Morris J., Jentoft M., Sorenson E., Butler G., Gastineau D., Dietz A., Windebank A.

Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration; 2016; 17 Supplement 1 (242).

ABSTRACT: Background: Mesenchymal stromal cells (MSCs) have been proposed to be a treatment for amyotrophic lateral sclerosis (ALS) due to their secretion of growth factors and alteration of the immune system. Preclinical studies have shown efficacy and human studies of bone-marrow derived MSCs have been promising. Methods: In order to determine the safety of intrathecal autologous adipose-derived MSC treatment for ALS, subjects with clinically-definite ALS by El Escorial criteria were enrolled into this Phase I dose escalation clinical trial. Doses ranged from 1×10(7) cells (single dose) to 1×10(8) cells (two monthly doses). Following intrathecal treatments, subjects underwent standardized follow-up, which included clinical examinations, revised ALS Functional Rating Scale (ALSFRS-R) questionnaire, blood and cerebrospinal fluid sampling, and magnetic resonance imaging (MRI) of the neuroaxis. Results: A total of 27 patients with ALS were enrolled and treated in this study. The safety profile was positive, with the most common side-effects reported being temporary low back and leg pain at the highest dose level. These clinical findings were associated with elevated cerebrospinal fluid protein and nucleated cells with MRI imaging of thickened lumbosacral nerve roots. Autopsies from four treated patients did not show evidence of tumor formation. Longitudinal ALSFRS-R questionnaires confirmed continued progression of disease in all treated patients. Conclusion: Intrathecal treatment of autologous adipose derived MSCs appears safe at the tested doses in ALS. These results warrant further exploration of efficacy in Phase II trials.

(2) Mesenchymal stem cells from adipose tissue in the treatment of amyotrophic lateral sclerosis – a case report.

Sekiya E.J., Jordy S.S., Kuhn T.I., Forte A., Bruniera G., Alves A., Bydlowski S.P

Cytotherapy (2013) 15:4 SUPPL. 1 (S33); April 2013

19th Annual Meeting of the International Society for Cellular Therapy; ISCT 2013 Auckland, New Zealand; 2013-04-22 to 2013-04-25

ABSTRACT: Amyotrophic Lateral Sclerosis (ALS) is a fatal neurodegenerative disease that selectively affects motor neurons in the brain and spinal cord leading to bulbar and respiratory muscle weakness. Currently there is no treatment proven effective and the disease is considered incurable. Mesenchymal Stem Cells (MSCs) derived from adipose tissue have immunomodulatory properties, inducing suppression and differentiation into neural cell types in vitro. Here we describe a compassionate intervention in a 66 years old male patient with Amyotrophic Lateral Sclerosis. The patient is suffering from Amyotrophic Lateral Sclerosis

diagnosed 4 years ago, unsuccessfully treated with Riluzole. It was observed atrophy of the shoulder girdle, upper limbs and thenar and hypothenar regions, fasciculations, tetra-pyramidal release (Hoffmann and Babinsky bilateral and global hyperreflexia) and inexhaustible clonus in the lower limbs. ALSFRS (ALS Functional Rating Scale) and its revised version ALSFRS-R were ALSFRS18 and ALSFRS-R26, respectively. After approval by the Ethics Committee, adipose tissue was collected from abdominal region by liposuction. Adipose tissue-derived MSCs were expanded under GMP conditions and administered intrathecally. Five days after the first infusion of 1×10(7) cells, the patient showed improvement of overall muscle strength and speech, exhaustible clonus in limbs, absence of Babinsky reflex in the lower left limb, and ALSFRS25 and ALSFRS-R33. One month later clinical condition worsened, with ALSFRS18 and ALSFRS-R26. The patient received other two higher dose infusions (5×10(7) and 10×10(7) cells) during the next 6 months and clinical data improved (ALSFRS25 and ALSFRS-R33) for a longer period. Moreover, no adverse effects were observed by MSC administration. In conclusion, although the study was performed in a single patient, intrathecal administration of autologous adipose tissue-derived MSC is potentially safe. It is tempting to speculate that administration of these cells could be beneficial for ALS patients, mainly using higher doses.

Diabetes

(1) Non-reconstructable peripheral vascular disease of the lower extremity in ten patients treated with adipose-derived stromal vascular fraction cells.

Carstens M.H., Gómez A., Cortés R., Turner E., Pérez C., Ocon M., Correa D.

Stem Cell Research (2017) 18 (14-21); 1 Jan 2017

ABSTRACT: We present a series of ten patients with peripheral vascular disease (PVD), secondary to arteriosclerosis (AS) and/or diabetes mellitus (DM), treated with local injection of non-expanded autologous, adipose-derived stromal vascular fraction (SVF) cells for the purposes of enhancing neovascularization and chronic wound healing. Adipose tissue was surgically harvested and processed to yield the heterogeneous SVF cells for immediate point-of-care injection. The gastrocnemius muscles and ulcers or wounds were locally injected with the resulting SVF. Response to treatment was evaluated both clinically based on pain-free ambulation, wound healing capacity over time and ankle/brachial index (ABI) measurements, and by imaging using MRI-based angiography. All patients exhibited clinical improvement (reduction in rest pain and claudication and improvements in ABI), with imaging signs of neovascularization in the majority (5 of 6) of patients in whom the evaluation was feasible. Similarly, 5 of 6 chronic wounds healed without further surgical intervention. This series highlights the utility of non-expanded adipose-derived heterogeneous SVF cell population processed at the point-of-care, to treat patients with end-stage PVD as an alternative to palliation or amputation.

(2) Expanded autologous adipose derived stem cell transplantation for type 2 diabetes mellitus: A preliminary report of 3 cases and review of literature.

Phuong T-B.L., Phuc V.P., Ngoc B.V., Loan T.-T.D., Ngoc K.P.

Biomedical Research and Therapy (2016) 3:12 (1034-1044); 15 Dec 2016

ABSTRACT: Type 2 diabetes mellitus (T2D) is the most common form of diabetes mellitus, accounting for 90% of diabetes mellitus in patients. At the present time, although T2D can be treated by various drugs and therapies using insulin replacement, reports have shown that complications including microvascular, macrovascular complications and therapy resistance can occur in patients on long term treatment. Stem cell therapy is regarded as a promising therapy for diabetes mellitus, including T2D. The aim of this study was to evaluate the safety and therapeutic effect of expanded autologous adipose derived stem cell (ADSC) transplantation for T2D treatment; the pilot study included 3 patients who were followed for 3 months. Methods: The ADSCs were isolated from stromal vascular fractions, harvested from the belly of the patient, and expanded for 21 days per previously published studies. Before transplantation, ADSCs were evaluated for endotoxin, mycoplasma contamination, and karyotype. All patients were transfused with ADSCs at 1-2x10(6) cells/kg of body weight. Patients were evaluated for criteria related to transplantation safety and therapeutic effects; these included: fever, blood glucose level before transplantation of ADSCs, and blood glucose level after transplantation (at 1, 2 and 3 months). Results: The results showed that all samples of ADSCs exhibited the MSC phenotype with stable karyotype (2n=46), there was no contamination of mycoplasma, and endotoxin levels were low (<0.25 EU/mL). No adverse effects were detected after 3 months of transplantation. Decreases of blood glucose levels were recorded in all patients. Conclusion: The findings from this initial study show that expanded autologous ADSCs may be a promising treatment for T2D.

(3) Safety and effect of adipose tissue-derived stem cell implantation in patients with critical limb ischemia: A pilot study.

Lee H.C., Park J.S., Choi J.H.

Global Heart, March 2014: 9:1 SUPPL. 1 (e11).

World Congress of Cardiology Scientific Sessions 2014, WCC 2014, Melbourne, VIC, Australia; 2014-05-04 to 2014-05-07

ABSTRACT: Introduction: Adipose tissue-derived mesenchymal stem cells (ADSCs) are potent and effective for inducing regeneration of damaged tissue. Treatment of critical limb ischemia (CLI) is occasionally difficult through bypass operation or percutaneous vascular intervention. Objectives: ADSC transplantation shows proangiogenic action in animal hindlimb ischemia models. We performed multiple intramuscular ADSC injections in CLI patients to treat diabetic foot and thromboangiitis obliterans (TAO) and assessed its safety and efficacy. Methods: The study included 15 CLI patients (15 men, median age: 52 years, range: 24-73 years) with ischemic resting pain in one limb with/without non-healing ulcers and necrotic foot (Rutherford class II-4:

n = 3, III-5: n = 9, III-6: n = 3) who did not undergo nonsurgical or surgical revascularization. ADSCs were isolated from adipose tissue of TAO patients (B-ADSCs), diabetes patients (D-ADSCs), and healthy donors (control ADSCs) by using 0.075% collagenase digestion. In a colony forming unit (CFU) assay using stromal vascular fraction, B-ADSCs and D-ADSCs yielded lesser colonies than normal ADSCs. Results: Culture-expanded B-ADSCs showed normal proliferation ability compared to control ADSCs. Adipogenic and osteogenic differentiation potential did not differ between B-ADSCs and control ADSCs. Culture-expanded D-ADSCs showed lower proliferation ability and osteogenic differentiation and higher adipogenic differentiation than control ADSCs. Multiple intramuscular ADSC injections cause no complications during the follow-up period (mean follow-up time: 6 months). Clinical improvement occurred in 66.7% of patients. Five patients required minor amputation during follow-up. All amputation sites healed completely. At 6 months, significant improvement was noted on pain rating scales and in claudication walking distance. Digital subtraction angiography (DSA) before and 6 months after ADSC implantation showed formation of numerous vascular collateral networks across affected arteries. Conclusion: Multiple intramuscular ADSC injections may be a safe alternative to achieve therapeutic angiogenesis in patients with CLI who are refractory to other treatment modalities.

(4) Autologous lipotransfer versus stromal vascular fraction enriched lipoinjection for diabetic foot wounds healing: a pilot study.

Journal of Biological Regulators and Homeostatic Agents, 2017, 31(4 Supplement 1), 141-146

Vicenti G, Solarino G, Pesce V, Moretti L, Notarnicola A, Carrozzo M, Rifino F, Moretti B.

ABSTRACT: Chronic ulcers of the lower limbs represent a significant social and economic burden. Diabetes is a strong risk factor for development of chronic lesions. Adult stem cells and growth factors derived from the adipose tissue are among the most promising therapeutic strategies for hard to heal wounds. Fat grafts have been used for several decades to treat soft tissue deformities, but despite its excellent characteristics, the outcome was unpredictable, due to partial necrosis and resorption of the graft. Stem cells' enrichment of these grafts or their injection into the edges of the ulcers have shown encouraging results in various experimental settings. In this pilot study, we compared the standard of care to autologous lipotransfer and stromal vascular fraction (SVF) enriched lipoinjection in 30 patients with diabetic foot ulcers, showing clear superiority of SVF enriched lipoinjection in terms of percentage of reduction of ulcers size and healing time.

WEB OF SCIENCE

Arthritis

(1) Intra-articular injection in the knee of adipose derived stromal cells (stromal vascular fraction) and platelet rich plasma for osteoarthritis.

Bansal H; Comella K; Leon J; Verma P; Agrawal D; Koka P; Ichim T.

Journal of Translational Medicine; June 2017 Volume: 15 Article Number: 141.

ABSTRACT: Stromal vascular fraction (SVF) can easily be obtained from a mini-lipoaspirate procedure of fat tissue and platelet rich plasma (PRP) can be obtained from peripheral blood. We evaluated the safety and preliminary efficacy of administering SVF and PRP intraarticularly into patients with osteoarthritis grade 1 and 2. Methods: A total of ten patients underwent a local tumescent liposuction procedure to remove approximately 100 ml of fat tissue from the abdomen. SVF was isolated using an enzyme digestion and re-suspended in PRP for intra-articular injection in the knee. The Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) score and six-minute walk distance (6MWD) were used to evaluate clinical effects and included measure of patient's subjective assessment of pain, joint mobility, and physical disability. WOMAC score, 6MWD and laboratory tests were repeated at 3 and 6 months and 1, 1.5 and 2 years. XRAY and MRI were completed at 1 year. Results: The average total WOMAC score was 64 at baseline and significantly reduced to 52 at 3 months, 46 at 6 months, 42 at 1 year, 38 at 1.5 years, and 41 at 2 years. Patients walked an average of 1310 feet at baseline and demonstrated a statistically significant improvement at 3 and 6 months and 1, 1.5, and 2 years post treatment. Cartilage thickness as determined by MRI improved by at least 0.2 mm in six patients, was unchanged in two patients and decreased by at least 0.2 mm in two patients. Conclusions: Overall, all the patients were pleased with the treatment results. They reported a reduction in pain levels, especially after 3 months. More importantly, the procedure demonstrated a strong safety profile with no severe adverse events or complications reported.

(2) Adipose Derived Stromal Cell (ADSC) Injections for Pain Management of Osteoarthritis in the Human Knee Joint.

Fodor, PB; Paulseth, SG.

Aesthetic Surgery Journal, Feb 2016; Volume: 36, Issue: 2, Pages: 229-236

ABSTACT: This safety and feasibility study used autologous adipose-derived stromal vascular cells (the stromal vascular fraction [SVF] of adipose tissue), to treat 8 osteoarthritic knees in 6 patients of grade I to III (K-L scale) with initial pain of 4 or greater on a 10-point Visual Analog Scale (VAS). Objectives: The primary objective of the study was evaluation of the safety of intra-articular injection of SVF. The secondary objective was to assess initial feasibility for reduction of pain in osteoarthritic knees. Methods: Adipose-derived SVF cells were obtained

through enzymatic disaggregation of lipoaspirate, resuspension in 3 mL of Lactated Ringer's Solution, and injection directly into the intra-articular space of the knee, with a mean of 14.1 million viable, nucleated SVF cells per knee. Metrics included monitoring of adverse events and preoperative to postoperative changes in the Western Ontario and McMaster Universities Arthritis Index (WOMAC), the VAS pain scale, range of motion (ROM), timed up-and-go (TUG), and MRI. Results: No infections, acute pain flares, or other adverse events were reported. At 3-months postoperative, there was a statistically significant improvement in WOMAC and VAS scores ($P < .02$ and $P < .001$, respectively), which was maintained at 1 year. Physical therapy measurements for ROM and TUG both improved from preoperative to 3-months postoperative. Standard MRI assessment from preoperative to 3-months postoperative showed no detectable structural differences. All patients attained full activity with decreased knee pain. Conclusions: Autologous SVF was shown to be safe and to present a new potential therapy for reduction of pain for osteoarthritis of the knee.

(3) Concentrated adipose tissue infusion for the treatment of knee osteoarthritis: clinical and histological observations.

Roato I, Belisario DC, Compagno M, Lena A, Bistolfi A, Maccari L, Mussano F, Genova T, Godio L, Perale, G.

International Orthopedics, Jan 2019, 1(43), 15-23

ABSTRACT: Osteoarthritis (OA) is characterized by articular cartilage degeneration and subchondral bone sclerosis. OA can benefit of non-surgical treatments with collagenase-isolated stromal vascular fraction (SVF) or cultured-expanded mesenchymal stem cells (ASCs). To avoid high manipulation of the lipoaspirate needed to obtain ASCs and SVF, we investigated whether articular infusions of autologous concentrated adipose tissue are an effective treatment for knee OA patients. Methods: The knee of 20 OA patients was intraarticularly injected with autologous concentrated adipose tissue, obtained after centrifugation of lipoaspirate. Patients' articular functionality and pain were evaluated by VAS and WOMAC scores at three, six and 18months from infusion. The osteogenic and chondrogenic ability of ASCs contained in the injected adipose tissue was studied in in vitro primary osteoblast and chondrocyte cell cultures, also plated on 3D-bone scaffold. Knee articular biopsies of patients previously treated with adipose tissue were analyzed. Immunohistochemistry (IHC) and scanning electron microscopy (SEM) were performed to detect cell differentiation and tissue regeneration. Results: The treatment resulted safe, and all patients reported an improvement in terms of pain reduction and increase of function. According to the osteogenic or chondrogenic stimulation, ASCs expressed alkaline phosphatase or aggrecan, respectively. The presence of a layer of newly formed tissue was visualized by IHC staining and SEM. The biopsy of previously treated knee joints showed new tissue formation, starting from the bone side of the osteochondral lesion. Conclusions: Overall our data indicate that adipose tissue infusion stimulates tissue regeneration and might be considered a safe treatment for knee OA.

(4) Limited evidence for adipose-derived stem cell therapy on the treatment of osteoarthritis.

Hurley ET, Yasui Y, Gianakos AL, Seow D, Shimozono Y, Kerkhoffs GMMJ, Kennedy JG.

Knee Surgery, Sports Traumatology, Arthroscopy Nov 2018; 26 (11): 3499-3507

ABSTRACT: The purpose of this systematic review is to evaluate the effects of adipose derived mesenchymal stem cells (ADSCs) in the treatment of osteoarthritis (OA) in the clinical setting. Methods: A literature search was performed in the MEDLINE, EMBASE, and The Cochrane Library Database up to January 2017 for inclusion and exclusion criteria. Criteria for inclusion were clinical studies demonstrating the effects of ADSCs on OA and written in English. The following variables were analyzed: donor site, volume of adipose tissue, preparation of ADSCs, clinical outcomes, and complication rate. Results: Sixteen studies (knee: 14 studies, multiple joints: 1 study, ankle: 1 study) were included in this systematic review. All of the studies prepared ADSCs in the form of the stromal vascular fraction (SVF). Inconsistencies between studies were found with regards to reported clinical variability, donor sites of SVF, and reported clinical outcomes. Nine studies used either platelet-rich plasma (PRP) (7/16) or fibrin (4/16) or both PRP and Fibrin (1/16), as an adjunct at time of SVF injection. All of the studies reported an improvement in clinical outcomes with the use of SVF. Five studies reported a 90% satisfaction rate, and no study reported any complications with liposuction. Five studies reported on complications, with a 5% incidence of swelling and pain. Conclusions: This systematic review demonstrated that ADSCs are currently used in the form of SVF. While SVF may produce favorable clinical outcomes with minimal risk of side effects on osteoarthritis, the variability in the data and the use of biological adjuvants have confounded the effectiveness of ADSCs. This study will help surgeons understand the limitations in the literature on ADSCs. Level of evidence Level IV, systematic review of level IV studies.

(5) Clinical results following intra-articular injection of adipose-derived stromal vascular fraction cells in patients with osteoarthritis of the knee.

Naomasa Yokota, Masayuki Yamakawa, Tomohiko Shirata, Tetsuya Kimura, Hideto Kaneshima.

Regenerative Therapy 6 (2017) 108e 112; http://dx.doi.org/10.1016/j.reth.2017.04.002

ABSTRACT: The purpose of this study was to evaluate the clinical results following intra-articular knee injection of Stromal Vascular Fraction (SVF cell therapy).
Methods: This study involved 13 consecutive patients who had received SVF cell therapy at our clinic before November 2015 and completed the 6-month post-treatment follow-up period. For each treatment, approximately 200 mL or more of subcutaneous adipose tissue was harvested using tumescent liposuction technique and manual aspiration of tissue from the lower abdomen using a suction cannula under local anesthesia in the operating room. The adipose tissue harvested was processed using the Celution Centrifuge IV in the cell processing room of our clinic. These cells were injected into the articular cavity of both knees directly. Outcome was assessed on the basis of patient questionnaires using VAS for pain, the Japanese Knee Osteoarthritis Measure (JKOM), the Western Ontario and McMaster Universities Osteoarthritis

Index (WOMAC). Results: The 13 patients (26 knee joints), consisting of 2 men and 11 women, had a mean age of 74.5± 5.4 years. Eleven patients (9 women, 2 men) presented Grade IV knee OA according to the KL classification. The remaining two patients, both women, had Grade III. Pre-operative scores of JKOM, WOMAC, VAS, and BS-POP (for patients) were 55.9 ± 21.0, 49.6 ± 20.4, 72.7 ± 18.2, and 18.5 ± 2.0. No serious adverse events were reported. One month after injection of SVF, all the scores of JKOM, WOMAC, and VAS were significantly improved over baseline ($P<0.01$). Ultimately, the scores were improved by an average of 35% over baseline for JKOM, 32% improvement in WOMAC, and 40% for pain (VAS). Conclusions: Our approach is unique in that it occurred within the context of the recently enacted Japanese Regenerative Medicine Safety Act which is the first in the world.

Spinal cord injury

(1) Intrathecal transplantation of autologous adipose-derived mesenchymal stem cells for treating spinal cord injury: A human trial.

Hur, JW, Cho, TH, Park, DH, Lee, JB Park, JY Chung, YG.

Journal of Spinal Cord Medicine; SN 1079-0268, 2016 (39) 6:655-664

ABSTRACT: Spinal cord injury (SCI) can cause irreversible damage to neural tissues. However, there is currently no effective treatment for SCI. The therapeutic potential of adipose-derived mesenchymal stem cells (ADMSCs) has been emerged. Objective: We evaluated the effects and safety of the intrathecal transplantation of autologous ADMSCs in patients with SCI. Participants/Interventions: Fourteen patients with SCI were enrolled (12 for ASIA A, 1 for B, and 1 for D; duration of impairments 3-28 months). Six patients were injured at cervical, 1 at cervico-thoracic, 6 at thoracic, and 1 at lumbar level. Autologous ADMSCs were isolated from lipo-aspirates of patients' subcutaneous fat tissue and 9 x 10(7) ADMSCs per patient were administered intrathecally through lumbar tapping. MRI, hematological parameters, electrophysiology studies, and ASIA motor/sensory scores were assessed before and after transplantation. Results: ASIA motor scores were improved in 5 patients at 8 months follow-up (1-2 grades at some myotomes). Voluntary anal contraction improvement was seen in 2 patients. ASIA sensory score recovery was seen in 10, although degeneration was seen in 1. In somatosensory evoked potential test, one patient showed median nerve improvement. There was no interval change of MRI between baseline and 8 months post-transplantation. Four adverse events were observed in three patients: urinary tract infection, headache, nausea, and vomiting. Conclusions: Over the 8 months of follow-up, intrathecal transplantation of autologous ADMSCs for SCI was free of serious adverse events, and several patients showed mild improvements in neurological function. Patient selection, dosage, and delivery method of ADMSCs should be investigated further.

Diabetes

(1) Accelerated healing of a diabetic foot ulcer using autologous stromal vascular fraction suspended in platelet-rich plasma.

Didangelos T., Koliakos G., Kouzi K., Arsos G., Kotzampassi K., Tziomalos K., Karamanos D., Hatzitolios A.

Regenerative Medicine: Vol: 13, Issue: 3, Pages: 277-281, DOI: 10.2217/rme-2017-0069, April 2018

ABSTRACT: We describe the case of a Type I diabetic patient with a refractory foot ulcer that remained unhealed for 2 years despite conventional therapy. Autologous adipose-derived stromal vascular fraction suspended in autologous platelet-rich plasma was applied to the wound, which completely healed within 1 month. The wound remained closed with no complications for a 2-year follow-up. Reporting of this and similar cases may lead to larger clinical trials that will prove the efficacy of this therapy that may offer accelerated healing and lessen the financial burden of more expensive therapeutic modalities.

(2) Co-infusion of insulin-secreting adipose tissue-derived mesenchymal stem cells and hematopoietic stem cells: novel approach to management of type 1 diabetes mellitus.

Thakkar, U. G.; Trivedi, H. L.; Vanikar, A. V.; et al.

International Journal of Diabetes in Developing Countries; Volume: 36, Issue: 4, Pages: 426-432, Dec 2016

ABSTRACT: Stem cell therapy (SCT) has promising results in regeneration of injured tissues/cells as well as correcting immune dysregulation. We present our experience of co-infusion of human adipose tissue-derived insulin-secreting mesenchymal stem cells (IS-AD-MSC) along with bone marrow-derived hematopoietic stem cells (BM-HSC) in type 1 diabetes mellitus (T1DM). This was an institutional review board-approved prospective non-randomized open-labeled clinical trial after informed consent of 20 patients (15 males and 5 females) with T1DM for SCT, with mean disease duration of 9 +/- 5.51 years. Their mean age and weight were 19.95 +/- 8.35 years and 49.9 +/- 14 kg, respectively. Our study includes T1DM with positive for glutamic acid decarboxylase (GAD) antibody and history of diabetic ketoacidosis (DKA). Generated IS-AD-MSC and BM-HSC were infused via femoral catheterization under local anesthesia into portal + thymic circulation and subcutaneous tissue with conditioning of injection rabbit anti-thymocyte globulin and Bortezomib. Patients were monitored for blood sugar, serum C-peptide, GAD antibodies, and glycosylated hemoglobin (Hb1Ac) at three monthly intervals post-therapy. Mean SC quantum infused 99.45 +/- 22.CD45-/90+ and CD45-/73+ were 47.22 and 24.66 %, respectively. Generated ISCs expressed transcription factors ISL-1, PAX-6, and IPF-1. Variable a5 mL, with mean 2.38 +/- 0.78 x 10(4) ISC/mu L, mean CD34+ 0.57 %, and mean sustained improvement in mean FBS, PPBS, HbA1c, and serum C-peptide was noted over a mean follow-up of 43.94 +/- 19.8 months with mean reduction of GAD antibody from 525.15 to 120.15 IU/mL. Mean insulin requirement decreased from 60.89 to 39.76 IU/day. There was

absence of DKA after SCT. No untoward effect/morbidity/mortality was recorded from SCT. Co-infusion of IS-AD-MSC with BM-HSC offers a safe and viable therapy for T1DM.

PUBMED

Arthritis

(1) Clinical results and second-look arthroscopic findings after treatment with adipose-derived stem cells for knee osteoarthritis.

Koh YG, Choi YJ, Kwon SK, Kim YS, Yeo JE.

Knee Surg Sports Traumatol Arthrosc. 2015 May;23(5):1308-16.

ABSTRACT: In the present study, the clinical outcomes and second-look arthroscopic findings of intra-articular injection of stem cells with arthroscopic lavage for treatment of elderly patients with knee osteoarthritis (OA) were evaluated. METHODS: Stem cell injections combined with arthroscopic lavage were administered to 30 elderly patients (≥65 years) with knee OA. Subcutaneous adipose tissue was harvested from both buttocks by liposuction. After stromal vascular fractions were isolated, a mean of 4.04 × 10(6) stem cells (9.7 % of 4.16 × 10(7) stromal vascular fraction cells) were prepared and injected in the selected knees of patients after arthroscopic lavage. Outcome measures included the Knee Injury and Osteoarthritis Outcome Scores, visual analog scale, and Lysholm score at preoperative and 3-, 12-, and 2-year follow-up visits. Sixteen patients underwent second-look arthroscopy. RESULTS: Almost all patients showed significant improvement in all clinical outcomes at the final follow-up examination. All clinical results significantly improved at 2-year follow-up compared to 12-month follow-up ($P < 0.05$). Among elderly patients aged >65 years, only five patients demonstrated worsening of Kellgren-Lawrence grade. On second-look arthroscopy, 87.5 % of elderly patients (14/16) improved or maintained cartilage status at least 2 years postoperatively. Moreover, none of the patients underwent total knee arthroplasty during this 2-year period. CONCLUSION: Adipose-derived stem cell therapy for elderly patients with knee OA was effective in cartilage healing, reducing pain, and improving function. Therefore, adipose-derived stem cell treatment appears to be a good option for OA treatment in elderly patients. LEVEL OF EVIDENCE: Therapeutic case series study.

(2) Autologous stromal vascular fraction therapy for rheumatoid arthritis: rationale and clinical safety.

Rodriguez JP, Murphy MP, Hong S, Madrigal M, March KL, Minev B, Harman RJ, Chen CS, Timmons RB, Marleau AM, Riordan NH.

Int. Arch Med. 2012 Feb 8;5:5. doi: 10.1186/1755-7682-5-5.

ABSTRACT: Advancements in rheumatoid arthritis (RA) treatment protocols and introduction of targeted biological therapies have markedly improved patient outcomes, despite this, up to 50% of patients still fail to achieve a significant clinical response. In veterinary medicine, stem cell therapy in the form of autologous stromal vascular fraction (SVF) is an accepted therapeutic modality for degenerative conditions with 80% improvement and no serious treatment associated adverse events reported. Clinical translation of SVF therapy relies on confirmation of veterinary findings in targeted patient populations. Here we describe the rationale and preclinical data supporting the use of autologous SVF in treatment of RA, as well as provide 1-, 3-, 6-, and 13-month safety outcomes in 13 RA patients treated with this approach.

(3) Adipose Mesenchymal Stromal Cell-Based Therapy for Severe Osteoarthritis of the Knee: A Phase I Dose-Escalation Trial.

Pers YM, Rackwitz L, Ferreira R, Pullig O, Delfour C, Barry F, Sensebe L, Casteilla L, Fleury S, Bourin P, Noël D, Canovas F, Cyteval C, Lisignoli G, Schrauth J, Haddad D, Domergue S, Noeth U, Jorgensen C; ADIPOA Consortium.

Stem Cells Transl Med. 2016 Jul;5(7):847-56. doi: 10.5966/sctm.2015-0245. Epub 2016 May 23.

ABSTRACT: Osteoarthritis (OA) is the most widespread musculoskeletal disorder in adults. It leads to cartilage damage associated with subchondral bone changes and synovial inflammation, causing pain and disability. The present study aimed at evaluating the safety of a dose-escalation protocol of intra-articular injected adipose-derived stromal cells (ASCs) in patients with knee OA, as well as clinical efficacy as secondary endpoint. A bi-centric, uncontrolled, open phase I clinical trial was conducted in France and Germany with regulatory agency approval for ASC expansion procedure in both countries. From April 2012 to December 2013, 18 consecutive patients with symptomatic and severe knee OA were treated with a single intra-articular injection of autologous ASCs. The study design consisted of three consecutive cohorts (six patients each) with dose escalation: low dose (2 × 10(6) cells), medium dose (10 × 10(6)), and high dose (50 × 10(6)). The primary outcome parameter was safety evaluated by recording adverse events throughout the trial, and secondary parameters were pain and function subscales of the Western Ontario and McMaster Universities Arthritis Index. After 6 months of follow-up, the procedure was found to be safe, and no serious adverse events were reported. Four patients experienced transient knee joint pain and swelling after local injection. Interestingly, patients treated with low-dose ASCs experienced significant improvements in pain levels and function compared with baseline. Our data suggest that the intra-articular injection of ASCs is a safe therapeutic alternative to treat severe knee OA patients. A placebo-controlled double-blind phase IIb study is being initiated to assess clinical and structural efficacy. SIGNIFICANCE: Although this phase I study included a limited number of patients without a placebo arm, it showed that local injection of autologous adipose-derived stem cells was safe and well tolerated in patients with knee osteoarthritis. This study also provides encouraging preliminary evidence of efficacy. Larger and

controlled long-term studies are now mandatory to confirm whether this new strategy of cell therapy can improve pain and induce structural benefit in osteoarthritis.

(4) Adipose-Derived Mesenchymal Stem Cells with Microfracture Versus Microfracture Alone: 2-Year Follow-up of a Prospective Randomized Trial.

Koh YG, Kwon OR, Kim YS, Choi YJ, Tak DH.

Arthroscopy. 2016 Jan;32(1):97-109. doi: 10.1016/j.arthro.2015.09.010.

ABSTRACT: To compare the clinical and radiologic efficacy of adipose-derived stem cells (ADSCs) with fibrin glue and micro-fracture (MFX) versus MFX alone in patients with symptomatic knee cartilage defects. METHODS: Patients who were aged 18 to 50 years and had a single International Cartilage Repair Society grade III/IV symptomatic cartilage defect (≥3 cm^2) on the femoral condyle were randomized to receive ADSCs with fibrin glue and MFX treatment (group 1, n = 40) or MFX treatment alone (group 2, n = 40). There was a lack of blinding for patients because of the additional intervention method (liposuction). The cartilage defect was diagnosed using preoperative magnetic resonance imaging (MRI), and quantitative and qualitative assessments of the repair tissue were carried out at 24 months by using the Magnetic Resonance Observation of Cartilage Repair Tissue scoring system with follow-up MRI. Clinical results were evaluated using the Lysholm score, the Knee Injury and Osteoarthritis Outcome Score (KOOS), and a 10-point visual analog scale for pain (0 points, no pain; 10 points, worst possible pain) preoperatively and postoperatively at 3 months, 12 months, and the last follow-up visit. RESULTS: The 2 groups had similar baseline patient characteristics. Follow-up MRI was performed at 24 months (mean, 24.3 months; range, 24.0 to 25.1 months) after the operation. Group 1 included 26 patients (65%) who had complete cartilage coverage of the lesion at follow-up compared with 18 patients (45%) in group 2. Significantly better signal intensity was observed for the repair tissue in group 1, with 32 patients (80%) having normal or nearly normal signal intensity (i.e., complete cartilage coverage of the lesion) compared with 28 patients (72.5%) in group 2. The mean clinical follow-up period was 27.4 months (range, 26 to 30 months). The improvements in the mean KOOS pain and symptom sub-scores were significantly greater at follow-up in group 1 than in group 2 (pain, 36.6 ± 11.9 in group 1 and 30.1 ± 14.7 in group 2 [P = .034]; symptoms, 32.3 ± 7.2 in group 1 and 27.8 ± 6.8 in group 2 [P = .005]). However, the improvements in the other sub-scores were not significantly different between group 1 and group 2 (activities of daily living, 38.5 ± 12.8 and 37.6 ± 12.9, respectively [P = .767]; sports and recreation, 33.9 ± 10.3 and 31.6 ± 11.0, respectively [P = .338]; quality of life, 38.4 ± 13.1 and 37.8 ± 12.0, respectively [P = .650]). Among the 80 patients, second-look arthroscopies were performed in 57 knees (30 in group 1 and 27 in group 2), and biopsy procedures were performed during these arthroscopies for 18 patients in group 1 and 16 patients in group 2. The second-look arthroscopies showed good repair tissue quality, although no significant intergroup difference was observed. The mean total histologic score was 1,054 for group 1 compared with 967 for group 2 (P = .036). Age, lesion size, duration of symptoms

before surgery, mechanism of injury, and combined procedures were not correlated with clinical results, Magnetic Resonance Observation of Cartilage Repair Tissue scores, and histologic outcomes at short-term follow-up. CONCLUSIONS: Compared with MFX alone, MFX and ADSCs with fibrin glue provided radiologic and KOOS pain and symptom sub-score improvements, with no differences in activity, sports, or quality-of-life sub-scores, in symptomatic single cartilage defects of the knee that were $3cm^2$ or larger, with similar structural repair tissue.

(5) Intra-articular injection of mesenchymal stem cells for the treatment of osteoarthritis of the knee: a proof-of-concept clinical trial.

Jo CH, Lee YG, Shin WH, Kim H, Chai JW, Jeong EC, Kim JE, Shim H, Shin JS, Shin IS, Ra JC, Oh S, Yoon KS.

Stem Cells. 2014 May;32(5):1254-66. doi: 10.1002/stem.1634.

ABSTRACT: Mesenchymal stem cells (MSCs) are known to have a potential for articular cartilage regeneration. However, most studies focused on focal cartilage defect through surgical implantation. For the treatment of generalized cartilage loss in osteoarthritis, an alternative delivery strategy would be more appropriate. The purpose of this study was to assess the safety and efficacy of intra-articular injection of autologous adipose tissue derived MSCs (AD-MSCs) for knee osteoarthritis. We enrolled 18 patients with osteoarthritis of the knee and injected AD MSCs into the knee. The phase I study consists of three dose-escalation cohorts; the low-dose (1.0 × 10(7) cells), mid-dose (5.0 × 10(7)), and high-dose (1.0 × 10(8)) group with three patients each. The phase II included nine patients receiving the high-dose. The primary outcomes were the safety and the Western Ontario and McMaster Universities Osteoarthritis index (WOMAC) at 6 months. Secondary outcomes included clinical, radiological, arthroscopic, and histological evaluations. There was no treatment-related adverse event. The WOMAC score improved at 6 months after injection in the high-dose group. The size of cartilage defect decreased while the volume of cartilage increased in the medial femoral and tibial condyles of the high-dose group. Arthroscopy showed that the size of cartilage defect decreased in the medial femoral and medial tibial condyles of the high-dose group. Histology demonstrated thick, hyaline-like cartilage regeneration. These results showed that intra-articular injection of 1.0 × 10(8) AD MSCs into the osteoarthritic knee improved function and pain of the knee joint without causing adverse events, and reduced cartilage defects by regeneration of hyaline-like articular cartilage.

(6) Infrapatellar fat pad-derived mesenchymal stem cell therapy for knee osteoarthritis.

Koh YG, Choi YJ.

Knee. 2012 Dec;19(6):902-7. Epub 2012 May 14.

ABSTRACT: The aim of the study was to determine if isolated mesenchymal stem cells (MSCs) derived from the infrapatellar fat pad could effectively improve clinical results when percutaneously injected into arthritic knees. LEVEL OF EVIDENCE: Therapeutic case-control study. METHODS: Twenty-five stem cell injections combined with arthroscopic debridement were administered to patients with knee OA. A mean of 1.89 × 10(6) stem cells were prepared with approximately 3.0 mL of platelet-rich plasma (PRP) and injected in the selected knees of patients in the study group. RESULTS: The mean Lysholm, Tegner activity scale, and VAS scores of patients in the study group improved significantly by the last follow-up visit. No major adverse events related to the injections were observed during the treatment and follow-up periods. The results were compared between the study and control groups, in which the patients had undergone arthroscopic debridement and PRP injection without stem cells. Although the preoperative mean Lysholm, Tegner activity scale, and VAS scores of the study group were significantly poorer than those of the control group, the clinical results at the last follow-up visit were similar and not significantly different between the two groups. CONCLUSIONS: The short-term results of our study are encouraging and demonstrate that infrapatellar fat pad-derived MSC therapy with intraarticular injections is safe and provides assistance in reducing pain and improving function in patients with knee OA.

(7) The use of stromal vascular fraction (SVF), platelet-rich plasma (PRP) and stem cells in the treatment of osteoarthritis: an overview of clinical trials.

Mehranfar S, Abdi Rad I, Mostafav E, Akbarzadeh A.

Artif Cells Nanomed Biotechnol; March 2019; 47(1):882-890.

ABSTRACT: Osteoarthritis (OA) is a major cause of disability across the world, which its prevalence is relatively high in elder population. Current accepted therapies such as exercise, anti-inflammatory drugs and intra-articular inoculation of corticosteroids are aimed at controlling symptoms in the affected patients. Surgical options including arthroplasty, osteotomy and joint replacement are other choices of treatment, which are invasive and can be applied in case of failure of conventional therapies. In the last few decades, efforts to treat musculoskeletal diseases are being increasingly focused on regenerative cellular therapies. Stromal vascular fraction (SVF), which obtained from adipose tissue, contains a variety of cells include mesenchymal stem cells (MSCs) and has shown to be effective in cartilage repair. Autologous blood products such as platelet-rich plasma (PRP) act as an adjuvant of surgical treatment and its intra-articular delivery has shown beneficial effects for OA treatment. Given the efficacy of such treatment

approaches in OA, this paper discusses both preclinical and clinical evidence with major focus on clinical trials.

(8) Stromal Vascular Fraction cell therapy for osteoarthritis in elderly: multicenter case-control study.

Michalek J, Vrablikova A, Darinskas, Lukac L, Prucha, J, Skopalik J, Travnik J, Cibulka M, Dudasova, Z.

J Clin Orthop Trauma, Jan 2019;10(1):76-80.

ABSTRACT: no abstract included; a cohort of 29 patients were treated with SVF with some improvement in pain scores.

(9) Stromal Vascular Fraction cells of adipose and connective tissue in people with osteoarthritis: a case control prospective, multi-centric non-randomized study.

Michalek J, Moster R, Lukac L, Proefrock K, Petrasovic M, Rubar J, Chaloupka A, Darinskas A, Kristek J, Travnik J, Jabandziev P, Cibulka M, Skopalik J, Kristkova Z, Dudasova, Z.

Global Surgery, May 2017, 3(3):1-9. Doi: 10.15761/GOS.1000163

ABSTRACT: Stromal vascular fraction (SVF), containing high amount of stem cells and other regenerative cells, can be easily obtained from loose connective tissue that is associated with adipose tissue. Here we evaluated safety and clinical efficacy of freshly isolated autologous SVF cells in a case control prospective multi-centric non-randomized study in patients with grade 2-4 degenerative osteoarthritis. Methods: A total of 1128 patients underwent standard liposuction under local anesthesia and SVF cells were isolated and prepared for application into 1-4 large joints. A total of 1856 joints, mainly knee and hip joints, were treated with a single dose of SVF cells. 1114 patients were followed for 12.1-54.3 months (median 17.2 months) for safety and efficacy. Modified KOOS/HOOS Clinical Score was used to evaluate clinical effect and was based on pain, non-steroid analgesic usage, limping, extent of joint movement, and joint stiffness evaluation before and at 3,6 and 12 months after the treatment. Results: No serious side effects, systemic infection or cancer was associated with SVF cell therapy. Most patients gradually improved 3-12 months after the treatment. At least 75% Score improvement was noticed in 63% of patients and at least 50% Score improvement was documented in 91% of patients 12 months after SVF cell therapy. Obesity and higher grade of OA were associated with slower healing. Conclusion: Here we report a novel and promising treatment approach for patients with degenerative osteoarthritis that is safe, cost-effective, and relying only on autologous cells.

Multiple Sclerosis

(1) Adipose-derived mesenchymal stem cells (AdMSC) for the treatment of secondary-progressive multiple sclerosis: A triple blinded, placebo controlled, randomized phase I/II safety and feasibility study.

Fernández O, Izquierdo G, Fernández V, Leyva L, Reyes V, Guerrero M, León A, Arnaiz C, Navarro G, Páramo MD, Cuesta A, Soria B, Hmadcha A, Pozo D, Fernandez-Montesinos R, Leal M, Ochotorena I, Gálvez P, Geniz MA, Barón FJ, Mata R, Medina C, Caparrós-Escudero C, Cardesa A, Cuende N, on behalf of the Research Group Study EudraCT.

PLoS One. 2018 May 16;13(5): e0195891. doi: 10.1371/journal.pone.0195891.

ABSTRACT: Currently available treatments for secondary progressive multiple sclerosis (SPMS) have limited efficacy and/or safety concerns. Adipose-mesenchymal derived stem cells (AdMSCs) represent a promising option and can be readily obtained using minimally invasive procedures. In this triple-blind, placebo-controlled study, cell samples were obtained from consenting patients by lipectomy and subsequently expanded. Patients were randomized to a single infusion of placebo, low-dose(1x106cells/kg) or high-dose(4x106cells/kg) autologous AdMSC product and followed for 12 months. Safety was monitored recording adverse events, laboratory parameters, vital signs and spirometry. Expanded disability status score (EDSS), magnetic-resonance-imaging, and other measures of possible treatment effects were also recorded. Thirty-four patients underwent lipectomy for AdMSCs collection, were randomized and thirty were infused (11 placebo, 10 low-dose and 9 high-dose); 4 randomized patients were not infused because of karyotype abnormalities in the cell product. Only one serious adverse event was observed in the treatment arms (urinary infection, considered not related to study treatment). No other safety parameters showed changes. Measures of treatment effect showed an inconclusive trend of efficacy. Infusion of autologous AdMSCs is safe and feasible in patients with SPMS. Larger studies and probably treatment at earlier phases would be needed to investigate the potential therapeutic benefit of this technique.

Diabetes

(1) Novel therapy for insulin-dependent diabetes mellitus: infusion of in vitro-generated insulin-secreting cells.

Dave SD, Vanikar AV, Trivedi HL, Thakkar UG, Gopal SC, Chandra T.

Clin Exp Med. 2015 Feb;15(1):41-5. doi: 10.1007/s10238-013-0266-1. Epub 2013 Dec 7

ABSTRACT: Insulin-dependent diabetes mellitus (IDDM) is a metabolic disease usually resulting from autoimmune-mediated β-cell destruction requiring lifetime exogenous insulin replacement. Mesenchymal stem cells (MSC) hold promise as a therapy. We present our experience of treating IDDM with co-infusion of in vitro autologous adipose tissue-derived MSC-differentiated insulin-secreting cells (ISC) with hematopoietic stem cells (HSC). This was an Institutional Review Board approved prospective non-randomized open-labeled clinical trial

after informed consent from ten patients. ISC were differentiated from autologous adipose tissue-derived MSC and were infused with bone marrow-derived HSC in portal, thymic circulation by mini-laparotomy and in subcutaneous circulation. Patients were monitored for blood sugar levels, serum C-peptide levels, glycosylated hemoglobin (Hb1Ac) and glutamic acid decarboxylase (GAD) antibodies. Insulin administration was made on sliding scale with an objective of maintaining FBS < 150 mg/dL and PPBS around 200 mg/dL. Mean 3.34 mL cell inoculums with 5.25 × 10(4) cells/μL were infused. No untoward effects were observed. Over a mean follow-up of 31.71 months, mean serum C-peptide of 0.22 ng/mL before infusion had sustained rise of 0.92 ng/mL with decreased exogenous insulin requirement from 63.9 international units (IU)/day to 38.6 IU/day. Improvement in mean Hb1Ac was observed from 10.99 to 6.72%. Mean GAD antibodies were positive in all patients with mean of 331.10 IU/mL, which decreased to mean of 123 IU/mL. Co-infusion of autologous ISC with HSC represents a viable novel therapeutic option for JDDM.

(2) Insulin-secreting adipose-derived mesenchymal stromal cells with bone marrow-derived hematopoietic stem cells from autologous and allogenic sources for type 1 diabetes mellitus.

Thakkar UG, Trivedi HL, Vanikar AV, Dave SD.

Presented at 2015 International Society for Cellular Therapy. Published by Elsevier Inc.

ABSTRACT: Stem cell therapy (SCT) is now the up-coming therapeutic modality for treatment of type 1 diabetes mellitus (T1DM). Methods: Our study was a prospective, open-labeled, two-armed trial for 10 T1DM patients in each arm of allogenic and autologous adipose-derived insulin-secreting mesenchymal stromal cells (IS-AD-MSC) + bone marrow-derived hematopoietic stem cell (BM-HSC) infusion. Group 1 received autologous SCT: nine male patients and one female patient; mean age, 20.2 years, disease duration 8.1 years; group 2 received allogenic SCT: six male patients and four female patients, mean age, 19.7 years and disease duration, 7.9 years. Glycosylated hemoglobin (HbA1c) was 10.99%; serum (S.) C-peptide, 0.22 ng/mL and insulin requirement, 63.9 IU/day in group 1; HbA1c was 11.93%, S.C-peptide, 0.028 ng/mL and insulin requirement, 57.55 IU/day in group 2. SCs were infused into the portal+thymic circulation and subcutaneous tissue under nonmyeloablative conditioning. Patients were monitored for blood sugar, S.C-peptide, glutamic acid decarboxylase antibodies and HbA1c at 3-month intervals. Results: Group 1 received mean SCs 103.14 mL with 2.65 ± 0.8 × 10(4) ISCs/kg body wt., CD34+ 0.81% and CD45-/90+/73+, 81.55%. Group 2 received mean SCs 95.33 mL with 2.07 ± 0.67 × 10(4) ISCs/kg body wt, CD34+ 0.32% and CD45-/90+/73+ 54.04%. No untoward effect was observed with sustained improvement in HbA1c and S.C-peptide in both groups with a decrease in glutamic acid decarboxylase antibodies and reduction in mean insulin requirement. SCT is a safe and viable treatment option for T1DM. Autologous IS-AD-MSC+ BM-HSC co-infusion offers better long-term control of hyperglycemia as compared with allogenic SCT.

Stroke

Search for "stroke" resulted in finding few reported investigations (shown below) related to the treatment of cardiomyopathy.

(1) Effects of the intramyocardial implantation of stromal vascular fraction in patients with chronic ischemic cardiomyopathy.

Comella K, Parcero J, Bansal H, Perez J, Lopez J, Agrawal A, Ichim T.

J Transl. Med. 2016 Jun 2;14(1):158. doi: 10.1186/s12967-016-0918-5

ABSTRACT: Stromal vascular fraction (SVF) can easily be obtained from a mini-lipoaspirate procedure of fat tissue. The SVF contains a mixture of cells including ADSCs and growth factors and has been depleted of the adipocyte (fat cell) population. We evaluated the safety and efficacy of administering SVF intramyocardially into patients with chronic ischemic cardiomyopathy. A total of 28 patients underwent a local tumescent liposuction procedure to remove approximately 60 ml of fat tissue. The fat was separated to isolate the SVF and the cells were delivered into the akinetic myocardial scar region using a trans-endocardial delivery system (MyoCath®) in patients who had experienced a previous myocardial infarct. The subjects were then monitored for adverse events, ejection fraction via echocardiogram and six-minute walk test (6MWT) over a period of 6 months. Results: The average EF was 29% at baseline and significantly increased to 35% at both 3 and 6 months. Patients walked an average of 349 m at baseline and demonstrated a statistically significant improvement at 3- and 6-months' post treatment of more than 80 m. Conclusions: Overall, patients were pleased with the treatment results. More importantly, the procedure demonstrated a strong safety profile with no severe adverse events or complications linked to the therapy.

(2) The Athena trials: Autologous adipose-derived regenerative cells for refractory chronic myocardial ischemia with left ventricular dysfunction.

Henry TD, Pepine CJ, Lambert CR, Traverse JH, Schatz R, Costa M, Povsic TJ, David Anderson R, Willerson JT, Kesten S, Perin EC.

Catheter Cardiovasc Interv, 2017 Feb 1;89(2):169-177. doi: 10.1002/ccd.26601. Epub 2016 Sep 23

ABSTRACT: To assess safety and feasibility of autologous adipose-derived regenerative cells (ADRCs), for treatment of chronic ischemic cardiomyopathy patients. Preclinical and early clinical trials suggest ADRCs have excellent potential for ischemic conditions. The Athena program consisted of two parallel, prospective, randomized (2:1, active: placebo), double-blind trials assessing intramyocardial (IM) ADRC delivery [40-million, n = 28 (ATHENA) and 80-million (ATHENA II) cells, n = 3]). Patients with an EF ≥20% but ≤45%, multivessel coronary artery disease (CAD) not amenable to revascularization, inducible ischemia, and symptoms of either angina (CCS II-IV) or heart failure (NYHA Class II-III) on maximal medical therapy were enrolled. All patients underwent fat harvest procedure (≤450 mL adipose), on-site cell processing

(Celution® System, Cytori Therapeutics), electromechanical mapping, and IM delivery of ADRCs or placebo. Results: Enrollment was terminated prematurely due to non-ADRC-related adverse events and subsequent prolonged enrollment time. Thirty-one patients (17-ADRCs, 14-placebo) mean age 65 ± 8 years, baseline LVEF (%) 31.1 ± 8.7 (ADRC), 31.8 ± 7.7 (placebo) were enrolled. Change in $V0_2$ max favored ADRCs (+45.4 ± 222 vs. -9.5 ± 137 mL/min) but there was no difference in left ventricular function or volumes. At 12-months, heart failure hospitalizations occurred in 2/17 (11.7%) [ADRC] and 3/14 (21.4%) [placebo]. Differences in NYHA and CCS classes favored ADRCs at 12-months with significant improvement in MLHFQ (-21.6 + 13.9 vs. -5.5 + 23.8, P = 0.038). Conclusions: A small volume fat harvest, automated local processing, and IM delivery of autologous ADRCs is feasible with suggestion of benefit in "no option" CAD patients. Although the sample size is limited, the findings support feasibility and scalability for treatment of ischemic cardiomyopathy with ADRCs.

Multiple Sclerosis

(1) Stem cell treatment for patients with autoimmune disease by systemic infusion of culture-expanded autologous adipose tissue derived mesenchymal stem cells.

Ra JC, Kang SK, Shin IS, Park HG, Joo SA, Kim JG, Kang BC, Lee YS, Nakama K, Piao M, Sohl B, Kurtz A.

J Transl Med. Oct 2011; 9:181. doi: 10.1186/1479-5876-9-181.

ABSTRACT: Prolonged life expectancy, life style and environmental changes have caused a changing disease pattern in developed countries towards an increase of degenerative and autoimmune diseases. Stem cells have become a promising tool for their treatment by promoting tissue repair and protection from immune-attack associated damage. Patient-derived autologous stem cells present a safe option for this treatment since these will not induce immune rejection and thus multiple treatments are possible without any risk for allogenic sensitization, which may arise from allogenic stem cell transplantations. Here we report the outcome of treatments with culture expanded human adipose-derived mesenchymal stem cells (hAdMSCs) of 10 patients with autoimmune associated tissue damage and exhausted therapeutic options, including autoimmune hearing loss, multiple sclerosis, polymyositis, atopic dermatitis and rheumatoid arthritis. For treatment, we developed a standardized culture-expansion protocol for hAdMSCs from minimal amounts of fat tissue, providing sufficient number of cells for repetitive injections. High expansion efficiencies were routinely achieved from autoimmune patients and from elderly donors without measurable loss in safety profile, genetic stability, vitality and differentiation potency, migration and homing characteristics. Although the conclusions that can be drawn from the compassionate use treatments in terms of therapeutic efficacy are only preliminary, the data provide convincing evidence for safety and therapeutic properties of systemically administered AdMSC in human patients with no other treatment options. The authors believe that ex-vivo-expanded autologous AdMSCs provide a promising alternative for treating autoimmune diseases.

Further clinical studies are needed that take into account the results obtained from case studies as those presented here.

Macular degeneration

(1) Preliminary study on electrophysiological changes after cellular autograft in age-related macular degeneration.

Limoli PG, Vingolo EM, Morales MU, Nebbioso M, Limoli C.

Medicine (Baltimore). 2014 Dec;93(29): e355. doi: 10.1097

ABSTRACT: Evolving atrophic macular degeneration represents at least 80% of all macular degenerations and is currently without a standardized care. Autologous fat transplantation efficacy was demonstrated by several studies, as these cells are able to produce growth factors. The aim of the work was to demonstrate possible therapeutic effect of the joined suprachoroidal graft of adipocytes, adipose-derived stem cells (ADSCs) in stromal vascular fractions (SVFs) of adipose tissue, and platelet-rich plasma (PRP). Twelve eyes in 12 dry age-related macular degeneration (AMD) patients, aged 71.25 (SD ± 6.8) between 62 and 80 years, were analyzed. A complete ocular evaluation was performed using best corrected visual acuity (BCVA), retinographic analysis, spectral-domain optical coherence tomography, microperimetry, computerized visual field, and standard electroretinogram (ERG). Each eye received a cell in graft between choroid and sclera of mature fat cells and ADSCs in SVF enriched with PRP by means of the variant second Limoli (Limoli retinal restoration technique [LRRT]). In order to test if the differences pre- and post-treatment were significant, the Wilcoxon signed-rank test has been performed. Adverse effects were not reported in the patients. After surgery with LRRT, the most significant increase in the ERG values was recorded by scotopic rod-ERG (answer coming from the rods), from 41.26 to 60.83 μV with an average increase of 47.44% highly significant ($P < 0.05$). Moderately significant was the one recorded by scotopic maximal ERG (answer coming from the rods and cones), from 112.22 to 129.68 μV with an average increase of 15.56% ($P < 0.1$). Cell-mediated therapy based on growth factors used appears interesting because it can improve the retinal functionality responses in the short term. The ERG could, therefore, be used to monitor the effect of cell-mediated regenerative therapies.

(2) Cell surgery and growth factors in dry age-related macular degeneration: visual prognosis and morphological study.

Limoli PG, Limoli C, Vingolo EM, Scalinci SZ, Nebbioso M.

Oncotarget. 2016 Jul 26;7(30):46913-46923. doi: 10.18632/oncotarget.10442.

ABSTRACT: The aim of this research was to study the overall restoration effect on residual retinal cells through surgically grafted autologous cells onto the surrounding tissue, choroid and retina in order to produce a constant secretion of growth factors (GFs) in dry age-related macular degeneration (AMD) patients. Thirty-six eyes of 25 patients (range 64-84 years of age) affected by dry AMD were included in study and divided in two groups by spectral domain-optical

coherence tomography (SD-OCT): group A with retinal thickness average (RTA) less than 250 microns (μm) and group B with RTA equal to or more than 250 μm. Adipocytes, adipose-derived stem cells from the stromal-vascular fraction, and platelets from platelet-rich plasma were implanted in the suprachoroidal space. Particularly, the following parameters were evaluated: best corrected visual acuity (BCVA) for far and near distance, retinal thickness maps, scotopic and photopic electroretinogram (ERG), and microperimetry (MY). All statistical analyses were performed with STATA 14.0 (Collage Station, Texas, USA). Results: 6 months after surgery, several values were statistically significant in the group with higher RTA. Also, patient compliance analysis (PCA) in relation to functional change perception appeared to be very good. Conclusions: The available set of GFs allowed biological retinal neuroenhancement. After 6 months it improved visual performance (VP), but the increase was better if RTA recorded by OCT was higher, probably in relation to the presence of areas with greater cellularity.

(3) Regenerative Therapy by Suprachoroidal Cell Autograft in Dry Age-related Macular Degeneration: Preliminary In Vivo Report.

Limoli PG, Vingolo EM, Limoli C, Scalinci SZ, Nebbioso M.

J Vis Exp. 2018 Feb 12;(132). doi: 10.3791/56469

ABSTRACT: This study is aimed at examining whether a suprachoroidal graft of autologous cells can improve best corrected visual acuity (BCVA) and responses to microperimetry (MY) in eyes affected by dry Age-related Macular Degeneration (AMD) over time through the production and secretion of growth factors (GFs) on surrounding tissue. Patients were randomly assigned to each study group. All patients were diagnosed with dry AMD and with BCVA equal to or greater than 1 logarithm of the minimum angle of resolution (logMAR). A suprachoroidal autologous graft by Limoli Retinal Restoration Technique (LRRT) was carried out on group A, which included 11 eyes from 11 patients. The technique was performed by implanting adipocytes, adipose-derived stem cells obtained from the stromal vascular fraction, and platelets from platelet-rich plasma in the suprachoroidal space. Conversely, group B, including 14 eyes of 14 patients, was used as a control group. For each patient, diagnosis was verified by confocal scanning laser ophthalmoscope and spectral domain-optical coherence tomography (SD-OCT). In group A, BCVA improved by 0.581 to 0.504 at 90 days and to 0.376 logMAR at 180 days (+32.20%) postoperatively. Furthermore, MY test increased by 11.44 dB to 12.59 dB at 180 days. The different cell types grafted behind the choroid were able to ensure constant GF secretion in the choroidal flow. Consequently, the results indicate that visual acuity (VA) in the grafted group can increase more than in the control group after six months.

(4) Stem Cell Therapies for Reversing Vision Loss.

Higuchi A, Kumar SS, Benelli G, Alarfaj AA, Munusamy MA, Umezawa A, Murugan K.

Trends Biotechnol. 2017 Nov;35(11):1102-1117. doi: 10.1016/j.tibtech.2017.06.016. Epub 2017 Jul 24

ABSTRACT: Current clinical trials that evaluate human pluripotent stem cell (hPSC)-based therapies predominantly target treating macular degeneration of the eyes because the eye is an isolated tissue that is naturally weakly immunogenic. Here, we discuss current bioengineering approaches and biomaterial usage in combination with stem cell therapy for macular degeneration disease treatment. Retinal pigment epithelium (RPE) differentiated from hPSCs is typically used in most clinical trials for treating patients, whereas bone marrow mononuclear cells (BMNCs) or mesenchymal stem cells (MSCs) are intravitreally transplanted, undifferentiated, into patient eyes. We also discuss reported negative effects of stem cell therapy, such as patients becoming blind following transplantation of adipose-derived stem cells, which are increasingly used by 'stem-cell clinics'.

(5) Vision Loss after Intravitreal Injection of Autologous "Stem Cells" for AMD.

Kuriyan AE, Albini TA, Townsend JH, Rodriguez M, Pandya HK, Leonard RE, Parrott MB, Rosenfeld PJ, Flynn HW Jr., Goldberg JL.

N Engl J Med. 2017 Mar 16;376(11):1047-1053. doi: 10.1056/NEJMoa1609583

ABSTRACT: Adipose tissue-derived "stem cells" have been increasingly used by "stem-cell clinics" in the United States and elsewhere to treat a variety of disorders. We evaluated three patients in whom severe bilateral visual loss developed after they received intravitreal injections of autologous adipose tissue-derived "stem cells" at one such clinic in the United States. In these three patients, the last documented visual acuity on the Snellen eye chart before the injection ranged from 20/30 to 20/200. The patients' severe visual loss after the injection was associated with ocular hypertension, hemorrhagic retinopathy, vitreous hemorrhage, combined traction and retinal detachment, or lens dislocation. After 1 year, the patients' visual acuity ranged from 20/200 to no light perception.

(6) Bilateral Retinal Detachments After Intravitreal Injection of Adipose-Derived "Stem Cells" in a Patient with Exudative Macular Degeneration.

Saraf SS, Cunningham MA, Kuriyan AE, Read SP, Rosenfeld PJ, Flynn HW Jr, Albini TA.

Ophthalmic Surg Lasers Imaging Retina. 2017 Sep 1;48(9):772-775. doi: 10.3928/23258160-20170829-16

A 77-year-old woman with exudative macular degeneration underwent bilateral intravitreal injections of "stem cells" at a clinic in Georgia. One month and 3 months after injection, she developed retinal detachments in the left and right eyes, respectively. Increased awareness within the medical community of such poor outcomes is critical so that clinics offering untested practices that have been shown to be potentially harmful to patients can be identified and brought under U.S. Food and Drug Administration oversight.

Chronic Obstructive Pulmonary Disease

(1) Autologous Stromal Vascular Fraction in the Intravenous Treatment of End-Stage Chronic Obstructive Pulmonary Disease: A Phase I Trial of Safety and Tolerability.

Comella K, Blas JAP, Ichim T, Lopez J, Limon J, Moreno RC.

J Clin Med Res. 2017 Aug;9(8):701-708. Epub 2017 Jul 1.

ABSTRACT: Chronic obstructive pulmonary disease (COPD) is a consistently progressive, ultimately fatal disease for which no treatment exists capable of either reversing or even interrupting its course. It afflicts more than 5% of the population in many countries, and it accordingly represents the third most frequent cause of death in the US, where it accounts for more than 600 billion in health care costs, morbidity, and mortality. Adipose tissue contains within its stromal compartment a high abundance of adipose stem/stromal cells (ASCs), which can be readily separated from the adipocyte population by methods which require less than 2h of processing time and yield a concentrated cellular preparation termed the stromal vascular fraction (SVF). The SVF contains all cellular elements of fat, excluding adipocytes. Recent clinical studies have begun to explore the feasibility and safety of the local injection or intravascular delivery of SVF or more purified populations of ASCs derived by culture protocols. Several pre-clinical studies have demonstrated a remarkable ability of ASC to nearly fully ameliorate the progress of emphysema due to cigarette smoke exposure as well as other causes. However, no prior clinical studies have evaluated the safety of administration of either ASC or SVF in subjects with COPD. We hypothesized that harvest, isolation, and immediate intravenous infusion of autologous SVF would be feasible and safe in subjects with COPD; and that such an approach, if ultimately determined to be efficacious as well as safe, would provide a highly practical method for treatment of COPD. METHODS: In this study, an initial phase I trial evaluating the early and delayed safety of SVF infusion was performed. Twelve subjects were enrolled in the study, in which adipose tissue was harvested using standard liposuction techniques, followed by SVF isolation and intravenous infusion of 150 - 300 million cells. Standardized questionnaires were administered to study feasibility as well as immediate and delayed outcomes and adverse events as primary endpoints. Secondary endpoints included subjective wellness and attitudes towards the procedure, as well as willingness to undergo the procedure a second time. The follow-up time ranged from 3 to 12 months, averaging 12 months. RESULTS: Of the 12 subjects, only one experienced an immediate adverse event, related to bruising from the liposuction. No observed pulmonary or cardiac issues were observed as related to the procedure. There were no deaths over the 12-month study period, and none identified in the subsequent telephonic follow-up. Attitudes toward the procedure were predominantly positive, and 92% of the study subjects expressed a desire to undergo the procedure a second time. CONCLUSIONS: This study is the first to demonstrate safety of SVF infusion in humans with serious pulmonary disease. Specifically, the use of intravenous infusion as a route to achieve pulmonary cellular targeting did not lead to clinical pulmonary compromise. The intravenous administration of SVF should be further explored as a potentially feasible and safe method for delivery leading to possible therapeutic benefit.

COCHRANE [CLINICAL TRIALS]

No additional completed trials have been identified. At the time of this search, there were some initial phase studies in various aforementioned conditions; these studies were either planned or recruiting as linked through clinicaltrials.gov.